The Journal of Parasitology

Volume 5 MARCH, 1919 Number 3

RECENT DISCOVERIES CONCERNING THE LIFE HISTORY OF *ASCARIS LUMBRICOIDES* *

B. H. Ransom and W. D. Foster†
Zoological Division, Bureau of Animal Industry, U. S. Department of Agriculture, Washington, D. C.

Ascaris lumbricoides is one of the most common and most important intestinal parasites of man. A roundworm sometimes known as *Ascaris suum,* or *A. suilla,* but morphologically indistinguishable from *A. lumbricoides* and probably of the same species, is of very frequent occurrence in the intestine of the pig. Until recently it had been generally assumed by parasitologists upon the basis of evidence collected by various investigators that the life cycle of *Ascaris lumbricoides* is simple and direct, that the eggs of the parasite which pass out of the intestine of the host animal in the feces, are swallowed by another human being or pig after a period of incubation sufficient for the development of the contained embryos to a vermiform stage, and that having been swallowed the eggs hatch in the alimentary tract, after which the embryos develop to maturity in the small intestine, the normal location of the adult worms. Stewart, however, in a series of notable papers (1916-1918) has lately presented the results of some investigations which have revealed imperfections in our former ideas of the life history of *Ascaris lumbricoides.* His contributions to our knowledge of this common parasite afford another striking illustration of the fact that prevailing and apparently well established views as to the life histories of parasites are often wrong.

Stewart first attempted to infect pigs by feeding them Ascaris eggs but failed. He then fed the eggs to rats and mice, and discovered that they hatched out in the alimentary tract, a fact already established by Davaine (1863), who also noted that newly hatched larvae could be found in the feces of rats soon after feeding the eggs. Stewart observed further, however, that not all of the young worms are thus eliminated in the feces. On the contrary many of them penetrate the

* Read at the meeting of the American Society of Zoologists, December 26, 1918.

† W. D. Foster died October 6, 1918.

intestinal wall and, aided by the circulation, migrate to the liver, spleen and lungs, and may be found in the liver and lungs two to four days after infection. He determined that the migrating worms undergo considerable growth and development, and in fact increase in length from about 0.22 mm. to as much as 1.4 mm. within a week after infection. They can be found in the bronchi and trachea seven or eight days after infection, later in the mouth, then in the esophagus, stomach and intestine. Having reached the intestine after their migrations through the lungs the larvae linger for a time in the cecum, but ultimately pass out of the body in the feces without development beyond the stage already reached in the lungs, except that they may become slightly larger reaching a maximum length of nearly 2.5 mm. According to Stewart, a rat or mouse may become quite free of the parasites as early as 16 days after infection. During the invasion of the lungs by the worms Stewart found that rats and mice commonly died from pneumonia. Influenced by his failure to infect pigs through feeding them Ascaris eggs and by his discovery of the behavior of the larval parasites in rats and mice, Stewart concluded that these animals act as intermediate hosts, the young worms being passed on to human beings and pigs through the contamination of food, water, etc., by the saliva or feces of rats or mice that had themselves become infected by swallowing the eggs of the parasite. It is necessary to admit that infection of man or pig in this way is theoretically possible, but it appeared to the writers following the publication of Stewart's earlier papers, that this explanation of the mode of infection was inadequate.

We had for a number of years been carrying on certain investigations relating to Ascaris in which infested pigs were utilized, and had repeatedly attempted to secure heavily infested subjects by feeding the animals with Ascaris eggs, but with very unsatisfactory results, so that our early experience with pigs was very similar to Stewart's. Nevertheless the results of Stewart's experiments with rats and mice and his failures and our failures to infect pigs did not seem necessarily to lead to the conclusion that rats and mice normally serve as intermediate hosts of *Ascaris lumbricoides*. The questions raised by Stewart's investigations were highly important from a practical as well as a purely scientific standpoint, and it appeared desirable that the Bureau of Animal Industry should collect further data that might assist in reaching definite conclusions. Accordingly the present writers repeated and supplemented Stewart's experiments. Without going into all the details of our work at this time, it may be stated that we have confirmed Stewart's results as to the behavior of Ascaris larvae in rats and mice. We have, however, in addition, made obser-

vations that appear to us to demonstrate very clearly that rats and mice are not normal intermediate hosts as Stewart suggested. In our opinion the real explanation of the behavior of Ascaris larvae in rats and mice is that the worms are merely going through the same course as they do in their usual hosts, man and pig. The only essential difference in the two instances is that in unsuitable hosts such as rats and mice the parasites are unable to complete their development to maturity, whereas in human beings and pigs after their migration through the lungs and return to the alimentary tract they can continue their growth to the adult stage. The value of Stewart's investigations, therefore, lies in the establishment of certain important facts relating to the migration of Ascaris larvae and not in the suggestions that he has made with reference to rats and mice as intermediate hosts.

In guinea-pigs and rabbits we have found that the larvae behave as they do in rats and mice with respect to their development, migration and elimination, and the fact that they are liable to cause a more or less serious pneumonia. From a young goat and a lamb after feeding them eggs of the pig Ascaris we have recovered immature worms that had developed beyond any stage yet obtained from rats, mice, guinea-pigs, or rabbits. In the case of the lamb, which two days after birth was fed Ascaris eggs and killed 103 days after feeding, we found in the intestine fifty partially grown ascarids, twelve males and thirty-eight females, the smallest male 60 mm., the largest female 130 mm., in length. The minimum lengths of the adults are about 150 mm. (male) and 200 mm. (female). The goat four days after birth was given a dose of Ascaris eggs, and 17 days later, a second dose. Seven days after the second dose the animal began to show symptoms of pneumonia and died three days later. In the lungs, trachea, esophagus and stomach numerous Ascaris larvae were found ranging from 1 to 2 mm. in length. These undoubtedly are traceable to the second feeding with Ascaris eggs, 10 days previously. In the small intestine were thousands of young ascarids measuring about 10 mm. in length, and these are traceable to the first feeding with eggs that took place 27 days before the death of the animal. These worms had developed to about four times the length of the largest that have been observed in experiments with the smaller laboratory animals.

From these experiments it is clear that the parasites behave in sheep and goats just as they do in rats, mice, guinea-pigs and rabbits, with the exception that after their return to the alimentary tract they are able to continue their development and approach the adult stage. These experiments also lend support to the common belief among parasitologists that the so-called *Ascaris ovis* occasionally found in sheep is merely the pig Ascaris in a strange host. It is of interest to

note that the specimens of *Ascaris ovis* whose measurements have been recorded are smaller than full grown specimens of *Ascaris lumbricoides,* and that fertile eggs appear never to have been seen. These are circumstances that are in accord with the results of our experiments and support the view that the sheep Ascaris is a parasite in the wrong host. Evidently the Ascaris of the pig is better adapted to existence in the sheep and goat than in rats, mice, guinea-pigs and rabbits. Apparently, however, it is unable to adapt itself sufficiently to reach the full measure of development attained in its usual host, the pig. In a scale of host adaptations we may therefore recognize three grades, rats, mice, guinea-pigs and rabbits in the lowest; sheep and goats in the intermediate grade, and pigs in the highest, with which we may also include human beings, if it be true that the Ascaris of man and of the pig are identical.

In our experiments on pigs we have found that the ingestion of Ascaris eggs by these animals is followed by the same series of phenomena that was observed in the experiments on other animals, including in some instances the occurrence of pneumonia. This has been noted in a previous paper (Ransom and Foster, 1917), and Stewart in one of his later articles (1918) has recorded the results of some experiments in which he observed the migration of Ascaris larvae through the lungs of pigs and the occurrence of pneumonia in these animals. Stewart, however, expressed himself as still unwilling to admit the development of Ascaris without an intermediate host. Owing to certain practical difficulties the experiments that we have thus far carried on with pigs as subjects have not been sufficiently controlled to exclude the possibility of the pigs themselves acting as their own intermediate hosts. That is, in all the cases in which we obtained intestinal infection with mature or nearly mature Ascaris following the feeding of the eggs to pigs, it is possible that the worms found had been reingested by the pigs after they had been passed out in the feces, continuation of their development beyond the lung stage having occurred only after such elimination and reingestion. So far, therefore, as our experiments on pigs are concerned, we cannot point to definite results disproving Stewart's views as to the necessity for an intermediate host. On the other hand no good evidence has yet been brought forth that Ascaris larvae after their migration through the lungs of an animal must necessarily pass out of the body and be reingested by the final host before they can develop to maturity.

There are certain important facts which have already been mentioned or alluded to by Lane (1917) and Low (1918) that are quite out of harmony with the hypothesis of the regular occurrence of the elimination and reingestion of Ascaris larvae as a necessity in the life

cycle. For example, the larvae after their migration through the lungs and elimination in the saliva or feces have very little resistance to unfavorable conditions, and though in moist media they can be kept alive for a time they quickly succumb to drying, a condition to which they are particularly liable to be exposed. The feeble resistance of the larvae after their passage through the lungs may be contrasted with the remarkable vitality of the eggs which have been kept alive for as long as five years, resist long periods of dryness, and are not killed by considerable periods of exposure to temperatures far below freezing. The egg stage is thus well adapted to withstand the hardships which the parasite must endure in its passage from one host to another, whereas the larvae that have passed through the lungs are not at all adapted to such an existence. On general principles it hardly seems probable that Ascaris could continue to exist if infection of the final host were brought about only by the ingestion of larvae which had already passed through the body of another animal and had been left exposed to the vicissitudes of the outer world in a feebly resistant condition.

As already stated, however, it may be admitted that infection in this way possibly sometimes occurs, though it has not yet been proved. On the other hand, the results of our experiments with the young lamb and the kid supply very strong proof of the correctness of the view that the final host becomes infected by swallowing properly incubated eggs and not by swallowing larvae that have passed through an intermediate host. In these experiments the very nature of the animals as well as their age practically excludes the possibility of infection through reingestion of larvae that had passed out of their bodies and dropped to the ground. In the light of the evidence from these experiments as well as that from the experiments with rats, mice, guinea-pigs, rabbits and pigs, and taking into consideration also the evidence which has been gathered by various investigators with reference to Ascaris infection of human beings, no other reasonable conclusion can be reached than that Ascaris has a direct life history without intermediate host, that infection occurs as a result of ingestion of the eggs, and that the larvae after migrating through the lungs return to the alimentary tract, settle down in the intestine if the animal is a suitable host and develop to maturity. It may be mentioned that very young pigs appear to be not only more susceptible to infection with Ascaris, but also more liable to develop pneumonia than other animals.

With reference to the production of pneumonia by Ascaris larvae it is of interest that Mosler (according to Leuckart, 1867) and Lutz (1888) observed lung symptoms in human beings a few days after the

ingestion of Ascaris eggs. In addition to the likelihood that Ascaris infection will be found to be responsible for certain lung troubles in human beings, particularly in children, it is quite likely that Ascaris has something to do with many of the cases of lung disease in pigs. Large numbers of young pigs suffer and die from lung affections the causes of which have never been satisfactorily explained. The symptoms shown by experimentally infected pigs at the time of the invasion of the lungs by the larvae are frequently exactly similar to those exhibited by pigs suffering from so-called "thumps," a popular name for a serious condition of very common occurrence among pigs, and it is accordingly not improbable that Ascaris is an important factor in the production of "thumps," especially when it is considered how very commonly Ascaris occurs as a parasite of pigs. Though we can not yet form a true estimate of the actual importance of Ascaris as a cause of lung disease it is evident that this parasite has capacities for harm not formerly suspected. Stewart's very interesting discovery of the migration of the larvae through the lungs has therefore not only added materially to our knowledge of the life history of Ascaris, but also by opening up a new line of investigation in pathology is likely to lead to a better understanding of the cause, prevention and treatment of certain diseases of the lungs.

The hatching of the eggs of Ascaris is an interesting question. It has been found by different investigators that when the eggs are swallowed hatching occurs in the small intestine. Hatching results not from any apparent digestion of the egg shell but from the active penetration of the shell by the contained embryo. Some writers have found that hatching will occur outside the body if the eggs are placed in certain solutions. We have been unable, however, to cause more than a very small percentage of the eggs to hatch outside the body in vitro. The factors which bring about the hatching of the eggs have not yet been determined. It is a noteworthy fact that if the eggs are injected beneath the skin of a guinea-pig they not only hatch, but that the larvae later appear in the lungs as they do following infection by way of the mouth, reaching a length of 0.5 mm. in seven days, and of 1.5 mm. in eleven days after the eggs are injected. Martin (1913) observed that the eggs of *Ascaris vitulorum* would hatch when introduced beneath the skin of a guinea-pig, but he did not follow the migrations of the larvae.

References Cited

Davaine, C. J. 1863.—Nouvelles recherches sur le développement et la propagation de l'ascaride lombricoide et du trichocéphale de l'home. Compt. rend. Soc. biol., Paris, (3) 4: 261-265.

Lane, Clayton. 1917.—*Ascaris lumbricoides* and coprophagia. Indian Med. Gaz., 52: 269-272.

1917.—Major Stewart on *Ascaris* infection. Indian Med. Gaz., 52: 301.

1917.—The life history of *Ascaris lumbricoides*. Indian Med. Gaz., 52: 380.

Leuckart, Rudolph. 1867.—Die menschlichen Parasiten und die von ihnen herrührenden Krankheiten. v. 2, 1. Lief.

Low, G. C. 1918.—The life-history of *Ascaris lumbricoides*. Brit. Med Jour., 1: 286.

Lutz, Adolph. 1888.—Zur Frage der Uebertragung des menschlichen Spulwurms. Weitere Mittheilungen. Centralbl. Bakter., 3: 425-428.

Martin, André. 1913.—Recherches sur les conditions du développement embryonnaire des nématodes parasites. Ann. sci. nat., zool., 18: 1-151.

Ransom, B. H., and Foster, W. D. 1917.—Life history of *Ascaris lumbricoides* and related forms. Jour. Agric. Research, 11: 395-398.

Stewart, F. H. 1916.—On the life history of *Ascaris lumbricoides*. Brit. Med. Jour., 2: 5-7.

1916.—The life-history of *Ascaris lumbricoides*. Brit. Med. Jour., 2: 474.

1916.—Further experiments on Ascaris infection. Brit. Med. Jour., 2: 486-488.

1916.—On the life-history of *Ascaris lumbricoides*. Brit. Med. Jour., 2: 753-754.

1917.—On the development of *Ascaris lumbricoides* Lin. and *Ascaris suilla* Duj. in the rat and mouse. Parasitology, 9: 213-227.

1917.—Note on *Ascaris* infection in man, the pig, rat, and mouse. Indian Med. Gaz., 52: 272-273.

1917.—The life-history of *Ascaris lumbricoides*. Indian Med. Gaz., 52: 379-380.

1918.—On the development of *Ascaris lumbricoides* and *A. mystax* in the mouse. Part 2. Parasitology, 10: 189-196.

1918.—On the life history of *Ascaris lumbricoides* L. Parasitology, 10: 197-205.

OBSERVATIONS ON AND EXPERIMENTS WITH *CUTEREBRA TENEBROSA* COQUILLET[1]

R. R. Parker and R. W. Wells

The studies carried on by the Montana State Board of Entomology in the Powder River Valley in 1916 afforded the writers opportunity for chance observations on the rodent bot fly, *Cuterebra tenebrosa* Coquillet. Among over a thousand rodents captured for examination as possible hosts of the Rocky Mountain spotted fever tick, *Dermacentor venustus* Banks, two were found, each infested with a single larva of this bot fly. The larvae were reared to adults and the following notes were made.

Record No. 1518, June 29. Found infesting breast of a pack rat (*Neotoma cinerea*). Air hole about three-sixteenths of an inch in diameter. Rat afterward died from trap injury and larva was squeezed out and placed on dirt. It immediately crawled in, the hole being left open. The fly emerged on August 25 after an interval of 47 days.[2]

Record No. 1526, July 19. Large bot larva found infesting grasshopper mouse (*Onychonomys lencogaster missouriensis*). It was embedded under the skin in front of the left hind leg. Larva emerged into bag in which mouse was placed for the ticks to crawl off. The adult emerged in Bozeman on Jan. 8, 1917, after an interval of 173 days.

On September 9, after returning to Bozeman, a living female of the fly was received in a box. The latter was left on the desk of the senior author for several days and when next examined was found to contain 186 eggs securely fastened to the pasteboard. They were not examined again until October 13, when the caps were removed from several eggs. Active larvae immediately crawled forth. This suggested the experimental infestation of rodents, and the experiments hereafter described were carried out. The larvae used were secured by removing the egg caps with a fine needle and gently assisting the larvae to make their escape. When the transfer was made to the mouth of the animal to be infested, the latter was made ready (prairie dogs were securely tied) and its mouth gently forced open and so held by a transverse pry. The cap of an egg was removed and the transfer made on the point of the needle as rapidly as possible and the animal released shortly thereafter.

---

1. Contribution from the Laboratory of the Montana State Board of Entomology, Bozeman, Montana.

2. This rat was also heavily infested with Siphonaptera and Anoplura as well as 45 larvae and nymphs of *Dermacentor venustus*.

Experiment 1. October 13. Four larvae transferred to the mouth of a prairie dog (*Cynomys ludovicianus*) and immediately disappeared. Three others were placed on a shaven spot on the neck, but it could not be seen that any effort was made to penetrate the skin. October 25, examination showed that 2 larvae had reached the subcutaneous tissue, one about at the middle of the left side (1), the other a little to the left of the middle beneath (2). Number 1 had punctured the skin and the larva was visible. It died about November 15, the abrasion healing quickly, leaving a hard lump that gradually disappeared. Number 2 was photographed on November 20 (Fig. 9) when the end could be seen very slightly protruding from the hole. It emerged during the following night and a photograph of the mature larva was made on November 21.

Experiment 2. This experiment was started with the hope that by using a considerable number of larvae, several being placed in the mouth at frequent intervals, that by subsequent dissection, the course of the larvae in the body might be traced. A prairie dog was again used as a host and 3 larvae were placed in its mouth on each of the following dates: October 27, 31, November 1, 2, 3, 4, 5, 6 and 7. A total of 27 larvae were used. On November 6 a small lump was noted on the belly, on November 7 this was slightly larger. On November 8 the host was killed and dissected, but no traces of the larvae were found either in the body cavity or tissues.

Experiment 3. Host, prairie dog. Three larvae were placed in its mouth on each of the following days, October 27 and 31, November 4 and 15. Larvae appeared under the skin as follows: (1) November 5 on right side, air hole noted on November 7; (2) November 16 on middle of neck dorsally; (3) November 17 slightly cephalad of (1) on right side; (4) November 17 slightly ventrad of (1) on right side; (5) November 17, on underside of left front thigh; (6) November 20, dorsal of middle of left side; (7) November 26 caudad and dorsad of left front leg.

On November 28, numbers 1, 4, 5 and 6 were dead, and numbers 2, 3 and 7 were still alive. Number 3 was removed, part of a cast skin coming with it. This was evidently the last molt and the chitin spangles had not yet become colored, the molt having apparently just taken place. The air hole was plugged at the time of removal. Numbers 4 and 7 emerged on December 13 and either crawled away or were destroyed by the dogs and were never recovered.

Experiment 4. Host, 13-striped ground squirrel (*Citellus tridecemlineatus pallidus*). Two larvae placed in its mouth on October 27 and 3 on November 21. Results negative.

Experiments 5, 6, 7 and 8. Hosts, Belgian hares. On November 8, 5, 4, 4 and 6, larvae were placed in the mouths of 4 Belgian hares, respectively. Results negative.

Experiment 9. Four larvae placed in the mouth of a prairie dog. December 14, 2 larvae appeared beneath the skin on the top of the fore shoulders. On January 8 one (1) was found to have emerged during the night, and on January 11 the second (2) was removed because of the condition of the host. This larva was located directly above the spine and was the only instance in which any of the prairie dogs gave evidence of being seriously affected by the presence of bot larvae. This dog had been used in one of the previous experiments. After removal the dog recovered rapidly. On January 8 number 1 was placed in a glass jar with 5 inches of dirt, on the 9th it had pupated about 2½ inches beneath the soil surface. Number 2 was placed in a similar jar on January 11, and after entering the ground voided considerable dark fluid from the anus. On January 12 it was still voiding similar excrementous matter. Pupation took place on January 14, but the insect died while in the pupal stage. On number 2, which was removed a little prematurely, the action of the larva in withdrawing and protruding the posterior spiracles indicated that the latter were protrusile before pupation. After pupation, however, they are

external as shown in figure 10. Number 1 had not emerged as an adult when the writers left Bozeman on March 25 for field work, and this record was never secured.

Exeriment 10. On December 18 two larvae were placed in the mouth of a house mouse (*Mus musculus*). The host was dissected on December 20, but no larvae were found.

Naturally our curiosity was aroused as to where the eggs were deposited under natural conditions, the conditions under which the operculum was displaced, the manner in which the larva gained entrance to the host and course followed by the larvae in reaching the subcutaneous tissues. The following points are presented for what they are worth. The egg cap when dry requires sufficient force to remove it to make it seem doubtful if the larva within can be instrumental in forcing it off. When eggs are moistened with saliva a dark outlining band appears around the margin of the cap (Fig. 1). Of several eggs placed intact in preserving fluid the caps of several were later found to be off and the larvae slightly to almost wholly protruding. Whether the removal of the cap was due to the action of the larva upon being irritated by the fluid, which must have penetrated the egg slowly, or was due to some action of the fluid on the egg, is uncertain. At least, however, it seems evident that the caps must have become loosened very soon after being placed in the fluid, since the larvae were able to crawl part way out of the egg before being overcome. It is also of interest that though the eggs were deposited within a period of a few days following September 9, 1916, yet larvae in the eggs not used in the experiments were still active the following March, about six months later, and had not escaped from the eggs.

When larvae were removed from the egg and placed upon a surface they would immediately attach themselves by an apparently sucker-like organ (Fig. 4) at the posterior end of the body and sway the body back and forth. The same habit was observed when placed on the skin of an animal. It is the recollection of the writers that these minute larvae were able to move about to some extent, very much after the manner of an inch worm. Unfortunately, no notes were made on this point. It is distinctly recalled that the larvae looped themselves in the manner above suggested, and that in one instance a larva, removed from the egg and placed beside it on the box, was afterwards found several inches away. What may be the value of the ability to attach themselves is not evident. The eggs were firmly "glued" to the box in which they were laid, indicating that they are fastened to something when deposited under natural conditions. The ventral surface of the egg is broad on the posterior two-thirds and the surface of the egg on this area is somewhat sunken inward. If

this is a groove, it is certainly very wide and shallow if intended to be attached to a single hair. The eggs were all fastened by this surface. After the cap was removed a delicate membrane was frequently noted covering the opening; this had to be broken before the larvae could escape.

It is possible, if other related bot flies were kept alive when captured, that eggs and larvae might be secured in a similar manner and valuable and suggestive information gained concerning life histories and habits.

SUMMARY OF DATA RELATING TO LIFE HISTORY

1. Under natural conditions the larvae of *Cuterebra tenebrosa* were found infesting pack rats and grasshopper mice. Prairie dogs were infested under laboratory conditions, but negative results were secured with Belgian hares and 13-striped ground squirrels.

2. A female deposited 186 eggs within a period of several days. These eggs contained active larvae which were still alive after six months in the laboratory.

3. By mechanically transferring larvae from eggs to the mouths of prairie dogs infestation was secured. In three experiments with these animals (Exp. 2 excluded because the host was killed) 20 larvae were used, of which 11 reached the subcutaneous tissue, 5 died in this situation and 6 emerged as fully matured larvae. (One of these was dissected out just as it was completing the last molt.)

4. Evidence that the larvae had reached the subcutaneous tissue was found on the twelfth day in two instances, and within maximum limits of 9 and 10 days in two other experiments.

5. The length of time elapsing after the first apparent evidence of larvae under the skin and before the skin was punctured was about two days.

6. The period spent in the subcutaneous tissue was 17, 25, 26 and 27 days in the several cases observed.

7. The total period from infestation to the emergence of the fully developed larva was respectively 37, 38 and 47 days in three instances.

8. After emergence from the host the mature larva entered the ground and soon pupated a few inches below the surface.

9. The period between the emergence of the mature larvae from the host and that of the fly was 47 days (June to August) in one instance and 173 days in another (July to January 8).

10. Winter apparently may be passed in the pupal stage.

11. Prairie dogs seemingly experienced no serious effects from the presence of the larvae even when several were present simultaneously. Infested dogs sometimes seemed less active and often appeared to favor the part of the body infested. In one experiment in which the bot was located above the spine on the fore shoulders the most serious effects were noted. When larvae died in the dogs the air holes healed quickly leaving lumps that gradually disappeared.

EXPLANATION OF PLATE

Figure 1. Eggs moistened with saliva showing dark band demarking the operculum.

Fig. 2. Normal egg.

Fig. 3. Egg with cap removed.

Fig. 4. Larva just removed from egg. Note sucker-like extension posteriorly.

Fig. 5. Larva just after emergence from host.

Fig. 6. Anterior end of mature larva.

Fig. 7. Posterior end of mature larva.

Fig. 8. Adult, female.

Fig. 9. Infested prairie dog just before emergence of larva.

Fig. 10. Puparium. Note spiracle posteriorly.

Fig. 11. Posterior spiracles of nearly mature larva with breathing apparatus retracted.

Fig. 12. Same, with breathing apparatus extruded.

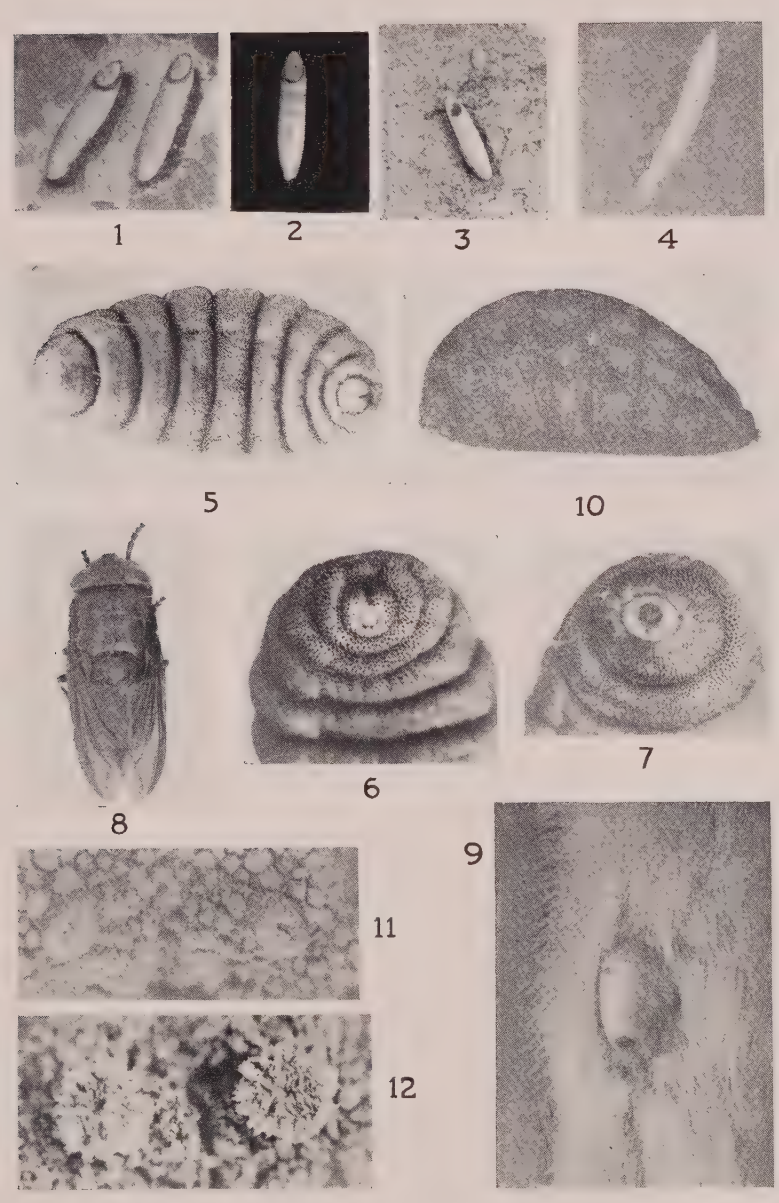


PLATE VIII

ON THE DEVELOPMENT OF *ASCARIS LUMBRICOIDES* L.

SADAO YOSHIDA
Pathological Department, Osaka Medical Academy, Osaka, Japan

Development of uninjured fertilized eggs. Leuckart reported that in the hot summer months the eggs may develop and form an embryo in two weeks. In my experiments 15 to 18 days was the minimum length of time required, and in the great majority of the eggs 30 days was necessary, even in the summer months. Widely varying stages of development are found in a single culture. The optimum temperature is 28 to 34° C., room temperature being favorable to rapid development during the summer.

Lutz, Epstein and others have proposed various means of producing an embryo within the shell. In the present experiments a sample of fresh feces mixed with water was put into a large test tube (or small beaker) and the contents stirred with a glass rod and filtered through a sheet of gauze. The sediment remaining in the test tube is mixed again with water and the filtration repeated. After this process is repeated two or three times most of the eggs will be found in the filtered fluid, from which they are collected by centrifugation. By removing the rubber plug from the bottom of a short tube the egg masses that accumulate are easily transferred into Petri dishes or other shallow vessels. A small quantity of aseptic moistened sand is added and the dish covered with a piece of gauze and a glass cover to protect from flies, and at the same time to allow free admission of air. This culture method is quick, easy and safe. The eggs can easily be separated from the sand because of the difference in specific gravity. The dishes are put into an incubator at 28 to 34° C. in the winter or left at room temperature in the summer months and must be kept moist. Exposure to strong direct sunlight checks development, and if continued, will kill the eggs.

When development is finished the full grown, coiled embryo within the shell is very active, especially in a warm temperature. Such eggs as these are said to be mature and are able to infect the body of the host when taken into the alimentary canal. Experiments were begun in May, 1916, and have been continued up to the present time save for interruptions due to accident.

Non-infectiveness of Unripe Eggs

That eggs in the various stages of development short of maturity do not develop into the embryo in the alimentary canal of the host, but are evacuated in the feces is proved by the following experiments.

Experiment I. May 15, 1916. Seven guinea-pigs were fed with eggs from feces evacuated on the preceding day which showed little or no signs of beginning division. Three days later three of the pigs were killed. No eggs were found in the stomach and very few in the small intestine, but a good many in the cecum and large intestine. None showed any evidences of development.

May 25, 1916. Two more of the pigs were killed. No eggs were found in the alimentary canal.

June 22, 1916 (thirty-seven days after feeding). The remaining two guinea-pigs were killed and examined. No eggs whatever were found.

Experiment II. July 30, 1917, 3 p. m., a rat was fed with eggs 14 days old. July 31, 9 a. m., fecal examination showed small quantities of eggs. Three hours later larger quantities were found. August 1, fecal examination negative. August 3, eggs in abundance in the feces. August 4, 8 a. m., eggs in abundance in feces; animal killed at 9 a. m.; the organs, especially liver and lungs, examined for larvae but with negative results. Liver and lungs both normal.

Experiment III. August 31, 1917, a rat was fed with eggs 15 days old. September 1, 9 a. m., fecal examination showed a small number of eggs. September 2, 8 a. m., animal died. A few eggs were found at autopsy in the intestine, but no embryos.

Experiment IV. September 2, 1917, 2 p. m., a rat was fed with unripe eggs. September 3, 8 a. m., fecal examination revealed no eggs. September 4, 8 a. m., abundant eggs in feces; 12 m., animal killed and the organs, especially liver and lungs, examined for larvae, but with negative results.

None of the eggs used in the foregoing experiments had reached the mobile embryo stage.

Hatching Place of Eggs

Both stomach and intestine have been suggested by various authors as possible hatching places of the ripe eggs in the animal or human host. I have carried out both incubation and feeding experiments to determine the hatching place of the eggs. Ripe eggs placed in 0.8 per cent. salt solution and incubated at 38° C. for 5 days did not hatch, nor did ripe eggs hatch when incubated for 6 days at 37 to 38° C. in gastric juice. The experiment was repeated with a 1 per cent. solution of sodium bicarbonate as medium with the same results.

Experiment V. August 3, 10 a. m., a mouse was fed with ripe eggs 59 days old. August 4, 8 a. m., numerous eggs were found present on fecal examination. 1 p. m. (twenty-seven hours after feeding) the mouse was killed. The stomach contained numerous eggs, one half hatched embryo, and three free embryos. The intestine contained three embryos and a good many eggs. Numerous eggs were found also in the cecum and large intestine.

Experiment VI. August 6, 4 p. m., two mice were fed with mature eggs 62 days old. August 7, 9 a. m. (seventeen hours after feeding) one mouse was killed. A small number of eggs were present in the stomach and small intestine, and numerous eggs in the cecum and large intestine, but no embryos. 11 a. m. (nineteen hours after feeding) the other mouse was killed. Eggs were found in the alimentary canal as in the first mouse, and in addition there were three embryos in the small intestine.

Experiment VII. August 3, 9 a. m., a guinea-pig wes fed with mature eggs 59 days old. August 4, 8 a. m., eggs were found in the feces; 10 a. m. (twenty-five hours after feeding) the animal was killed. A few eggs were found in the

stomach and large intestine, but none in the small intestine. No embryos were found.

Experiment VIII. August 20, 2 p. m., ripe eggs 35 days old were given to a guinea-pig. August 21, 9 a. m. (nineteen hours after feeding) the animal was killed. A few eggs were found in the stomach and small intestine, some in the large intestine. No embryos were found.

Experiment IX. August 20, noon, ripe eggs 35 days old were fed to a rat with biscuits. The animal did not eat all of the biscuits until the next day. August 22, 10 a. m. (forty-six hours after feeding) the rat was killed. A few eggs were present in the stomach and lower part of small intestine. In the cecum there were three free dead embryos and a half hatched embryo, as well as numerous eggs, and in the upper part of the large intestine there were two free dead embryos and abundant eggs.

Experiment X. August 24, 4 p. m., three mice were fed with ripe eggs 35 days old. August 25, 9 a. m. (seventeen hours after feeding) one mouse was killed. A few eggs were found in the stomach and small intestine, and they were numerous in the cecum and large intestine, but there were no embryos; 12 noon (twenty hours after feeding) another of the mice was killed. The findings were similar to those in the first mouse. August 30, 11 a. m. (six days after feeding), the remaining mouse was killed. There were no eggs in the alimentary canal. The liver and lungs were normal.

Apparently the eggs used in Experiment X did not have sufficient life to develop. This was probably due to the fact that the culture from which the eggs was taken, together with other cultures, was exposed on August 24 to strong direct sunlight in an attempt to determine whether the sunlight would accelerate development. The contrary was thus proved to be true.

It is impossible to draw a conclusion from the six foregoing experiments with regard to the hatching place of the eggs, though the evidence, combined with that of previous investigators, favors the small intestine.

The average time of retention of eggs in the animals appears from our records of these experiments to be from 2 to 5 and usually 3 days. There were always some mature eggs among the greater number of immature eggs evacuated. It seems reasonable to assume that these apparently fully matured and healthy eggs were internally injured in such a way as to make further development impossible. It is clear that not all the mature eggs develop, especially in the case of a young culture. To obtain a heavy infection with Ascaris larvae it is necessary to use large quantities as completely developed as possible (40 days or more). The failure of many investigators to observe development of Ascaris within the animal host was perhaps due to lack of recognition of this point.

Migration of Larvae in the Body of Host

From the foregoing experiments, and from those of previous investigators, it seemed probable that the ripe eggs of Ascaris hatch in the

intestine of the animal within 12 to 19 hours, after which the larvae migrate into the other organs, particularly the liver and lungs. The following experiments demonstrate this latter point.

Experiment XI. June 23, 1916, two guinea-pigs were fed with mature eggs 40 days old. June 26 (seventy-seven hours after feeding) one pig was killed by mistake. Two small nematode larvae were found in the liver which afterwards proved to be young Ascaris. The remaining pig was killed June 30. It was impossible to find any nematode larvae in the liver. On opening the pleural cavity, however, the lungs were found to show heavy hemorrhage and to contain two small nematode larvae a little larger than those found in the liver in the preceding experiment.

Experiment XII. August 8, 1917, ripe eggs 66 days old were fed to a guinea-pig. Three days later the pig was killed. The liver, which was normal in color and in size, contained eight larvae. The lungs were normal.

Experiment XIII. August 3 and 4, a guinea-pig was fed with ripe eggs 59 and 60 days old, respectively. The animal was killed on August 7. The liver and lungs were externally normal. Eighteen larvae were found in the former.

Experiment XIV. August 10, a guinea-pig was fed with ripe eggs 66 days old. The pig was killed August 14. The liver was apparently normal, though somewhat lighter in color, and contained twelve larvae. The lungs were normal except for slight reddish spots and contained three of the worms.

Experiment XV. August 29, a guinea-pig was fed with mature eggs incubated in artificial gastric juice at 37 to 38° C. for six days, and also with some incubated in a 1 per cent. solution of sodium bicarbonate at the same temperature for five days. On September 3 the animal was killed. The liver was apparently normal, but faintly hemorrhagic and slightly lighter in color. It contained sixteen larvae. The lungs were somewhat spotted and contained nine of the worms.

Experiment XVI. July 24, a quantity of mature eggs 49 days old were given to a guinea-pig. Feeding with the eggs from the same lot was repeated on July 27. On August 1 the animal was killed. The surface of the lungs was covered with reddish or dark spots due to new and old hemorrhages, and fourteen specimens were obtained from it. Two larvae were found in the liver, one in the trachea and one in the small intestine (dead).

Experiment XVII. A guinea-pig was fed with a small quantity of feces containing ripe eggs and with mature eggs from a culture 56 days old on July 31, and again on August 1. The animal was killed on August 8. The lung surfaces were covered with hemorrhagic spots, and sixty-four specimens of larvae were collected from them. The other organs gave negative results.

Experiment XVIII. July 24, ripe eggs 49 days old were given to two young rats with food, which was not all eaten until the next day. The rats were put into separate cages on July 27, and rat A was fed with a large dose of ripe eggs. Rat B was killed on July 30. The liver only yielded specimens of the worms, two being contained in it. Rat A was killed August 2. The lungs were somewhat spotted with hemorrhages and contained eight worms. The other organs were negative.

The foregoing experiments indicate that the completely matured eggs hatch in the intestine, whence the larvae migrate to the liver, lungs, and trachea, returning finally through the pharynx to the intestines. The liver is apparently not affected by the larvae, but the lungs usually show considerable hemorrhage.

The Fate of the Larvae in the Body of the Feeding Animal

The larvae finally reaching the intestine in these feeding animals are apparently not able to develop into adult form there, being voided with the feces within a few days, presumably because the animals are not the normal host of the parasite. Hence, it seemed reasonable to suppose that completely developed larvae from the lungs or trachea of feeding animals, when ingested by the human being, will grow into adult form in the human intestine. To determine this point 35 specimens of larvae from the lungs of a guinea-pig were taken by the writer on August 8. The details of the experiment will be described later.

The following several experiments were made to determine the period during which the larvae appear in the various organs of the feeding animals.

Experiment XIX. October 11, 1917, two guinea-pigs were fed with ripe eggs, 42 days old. Twenty-four hours later guinea-pig A was killed. Embryos were not found in any part of the alimentary canal or even in the abdominal cavity, and only a few in the stomach and intestine. Guinea-pig B was killed on October 19. Six large larvae (2 mm. in length) were found in the left lung, which was severely hemorrhaged, and three in the trachea.

Experiment XX. November 27, six guinea-pigs were fed with ripe eggs, 68 days old. Guinea-pig A was killed on the next day, and two living larvae were found in the large intestine, but none in other organs. Animal C was found dead on December 3. The liver was slightly infected, but the lungs were heavily hemorrhaged and consolidated. The upper part of the trachea contained two larvae. Animal D was found dead on December 5. The liver was normal. The lungs were dark brown, hemorrhagic, and contained numerous larvae. A few larvae were found in the trachea. Animal E was killed on December 9. The lungs were extremely hemorrhagic, consolidated and dark brown in color, but contained no larvae. Two larvae were found in the trachea and five dead ones in fecal pellets from the rectum. Animal F was killed on December 10. Lungs: As in the preceding animal. One specimen was found in the lung, and three in the large intestine, but none in the trachea.

Experiment XXI. December 12, two guinea-pigs were fed with ripe eggs, 85 days old. Animal A was found dead on December 14. The liver was heavily infected with the larvae. The abdominal cavity was free from the larvae. Guinea-pig B was killed on December 15. The liver was severely infected. The lungs were slightly hemorrhagic and contained a few larvae.

Experiment XXII. October 29, ripe eggs, 60 days old, were given to two guinea-pigs, one of which was found dead on November 1 (animal A). The liver was severely infected with larvae. The other organs were free. Animal B was killed on November 7. The liver was normal. The lungs were spotted with dark brown color, and the anterior lobe of the left lung was consolidated. Numerous larvae were found in the lungs and trachea, one dead specimen in the small intestine, and four living and one dead larva in the large intestine. The trachea contained three specimens in the upper portion, twenty-four in the middle portion and fifty-one in the lower. Fifty of these completely developed living specimens were later ingested by the writer. They varied from 1.23 to 1.62 mm. in length.

Experiment XXIII. November 10, two guinea-pigs were fed with ripe eggs, 54 days old (to A) and 72 days old (to B). B was killed November 14. The liver was heavily infected, and the lungs slightly hemorrhagic and apparently consolidated. Three specimens of larvae were found in the heart. Animal A was found dead on November 25. The outer surface of the lungs was light red and the inner surface dark red in color. They were hemorrhagic and consolidated. Three specimens were present in the left lung, six in the trachea, which contained a small quantity of bloody mucus, and two dead larvae in the large intestine.

Experiment XXIV. November 15, a guinea-pig (A) was fed with ripe eggs, 57 days old, cultivated in a 0.5 per cent. solution of hydrochloric acid, and another (B) with eggs of the same age cultivated in a 0.2 per cent. solution. A was found dead on the 20th. The liver was infected with larvae, and the lungs were hemorrhagic, consolidated and contaminated with numerous larvae. Animal B was found dead on November 22. The liver was slightly infected. The lungs were severely hemorrhaged and apparently consolidated. Abundant larvae of small size were found in the organ. The trachea contained a small quantity of bloody mucus in which numerous larvae were found.

Experiment XXV. November 17, two guinea-pigs were fed with fully matured eggs, 59 days old cultivated in a 0.1 per cent. solution of carbonic acid. A was found dead in the morning of the 22d. The larvae were found in the liver and lungs, and the latter was dark brown in color from heavy hemorrhage. The trachea contained a small quantity of bloody mucus in which three small larvae were present. Animal B was found dead November 26. The liver was free from the larvae. The lungs were dark brown from hemorrhage, consolidated, and contained numberous larvae of various sizes. There were also larvae in the trachea.

Experiment XXVI. November 8, guinea-pig A was fed with ripe eggs, 58 days old, and B with ripe eggs cultivated in a 0.3 per cent. solution of carbonic acid for seventy days. C received mature eggs 70 days old. Animal A was killed on the 13th. The liver and lungs were infected, the trachea free. The lungs contained light and dark red hemorrhagic spots. Two larvae were found in the heart. B was found dead on November 17. The lungs were dark brown with light red margin and were somewhat infected with larvae. Numerous larvae were found in the trachea and six in the large intestine. C was found dead on November 19, and eight large specimens (1.75 to 1.91 mm. long) were found in fecal pellets from the rectum.

Experiment XXVII. November 16, two guinea-pigs were fed with eggs 58 days old. Animal A was found dead on the 24th. The liver contained a few worms. The lungs showed heavy hemorrhage and contained numerous specimens of the worm. The trachea contained some blood and two larvae. B was killed on November 26. The lungs showed severe hemorrhage, were consolidated, and contained a few larvae. The trachea and large intestine also contained a few.

Experiment XXVIII. October 20, two guinea-pigs were fed with eggs 51 days old. Animal A was lost; B was found dead on the 29th. The liver was free. The lungs were somewhat hemorrhagic and contained numerous larvae.

Experiment XXIX. October 20, a guinea-pig was fed with ripe eggs cultivated in a 0.1 per cent. solution of carbonic acid for 51 days. It was found dead on the 29th. The lungs were severely hemorrhaged and consolidated, and large larvae were abundant in the lungs and trachea. Three living larvae were also found in the large intestine.

Summary of Experiments XIX to XXIX

The larvae first appear in the liver on the second day (about 44 hours after feeding), and they are present until the sixth or seventh day, being most abundant on the third, fourth and fifth days, and disappearing after the eighth day.

As Experiments XXI and XXIII show, the larvae are first found in the lungs on the third or fourth day, and a few persist even to the fourteenth or fifteenth day after feeding. They are most abundant from the sixth to the eighth day.

In the trachea the larvae appear first on the fifth or six day and persist as long as they are present in the lungs, being most abundant on the eighth and ninth days. From the trachea they migrate into the mouth and thence down the alimentary tract of the host. The appearance of larvae in the alimentary tract begins after the eighth day. They are rarely found in the esophagus, stomach and small intestine, apparently passing through rapidly, but accumulate in the cecum and large intestine, where some of them are found dead. All are sooner or later voided in the feces.

Severely infected lungs are almost always hemorrhagic, consolidated, and dark brown in color. Epistaxis was not encountered in the animals of these experiments, but Stewart has reported its frequent occurrence in the infected rat.

Experiments on Other Mammals

Further experiments with regard to migration were made in other mammals and in the human body. For this purpose the rabbit, cat and monkey were used.

Experiment XXX. January 22, 1918, two monkeys were fed with large quantities of ripe eggs 76 and 87 days old. A was again fed with a large dose of eggs, 91 days old, on the 26th. B was found dead on the 29th, and Ascaris larvae were found in the lungs. Monkey A was killed on February 1. No larvae were found in the liver. The lungs were heavily infected. Two larvae were found in the trachea, large quantities in the stomach, and a few in the small intestine. The lungs were slightly hemorrhagic and consolidated.

Experiment XXXI. March 9, a cat was fed with eggs 131 days old. March 11, there was another feeding from eggs of the same lot. The animal was killed on the 18th. The lungs were slightly hemorrhagic and infected.

Experiment XXXII. A rabbit was thrice fed with Ascaris eggs, on December 29, 1917; January 21, and March 9, 1918. On March 19 the rabbit was killed. The lungs were slightly hemorrhagic and infected with numerous larvae. A few larvae were present in the trachea and small intestine.

From these experiments it is concluded that the migration of Ascaris larvae is definite in any feeding animal in which the larvae are alive. It was therefore to be supposed that they follow the same course in the human body as in the feeding animals. Personal experiments later confirmed this belief.

Experiments on Immunity

The next point to determine was whether or not the infected animal requires any protection against a second infection with Ascaris larvae.

Experiment XXXII. The intervals between the first and third and between the second and third feedings were seventy and forty-seven days, respectively. The rabbit was killed on the tenth day after the feeding, and its lungs, trachea and small intestine were infected with the larvae.

Experiment XXXIII. January 21, a guinea-pig was fed with ripe eggs 81 days old. March 7, it was again fed with eggs 91 days old. The animal was killed on March 14, seven days after the last feeding. The liver and lungs were slightly infected and the latter somewhat hemorrhagic.

The animals in these two experiments, having recovered from the earlier infections, apparently had no protection against subsequent ones.

Experiment XXXIV. January 23, a guinea-pig was fed with eggs 78 days old; March 7 the feeding was repeated with eggs 129 days old. It was killed on March 18. No larvae were found.

Experiment XXXV. Two successive feedings of eggs 129 days old were given on March 7 and March 9. When it was killed on the 27th, eight days after the last feeding, it was found to be uninfected.

The results of the two last experiments might be ascribed either to the presence of an immunity acquired by the previous infection or to the inability of the eggs used in the second feeding to develop, the eggs having been in dry condition for some days. Stewart has reported a case in which the rat was immunized by one infection with Ascaris larvae. Prof. Fujinami reported the immunization of a horse by one heavy infection with *Sch. japonicum*.

The Migrating Power of the Larvae

Experiment XXXVI. On March 16 Ascaris larvae from the liver of a guinea-pig killed sixty-seven hours after feeding were injected into the abdominal cavity of two guinea-pigs. Guinea-pig A was killed March 23. The lungs were slightly hemorrhagic and infected. B was killed on the following day. The condition of the lungs was the same as in A. The small portions of the middle lobes on both sides were dark gray in color and evidently consolidated.

It was not determined in this experiment by what route the injected larvae migrate into the lungs — by the blood vessel after penetrating the liver, or by penetrating the lungs from the surface after passing through the diaphragm. The experiment gives clear evidence, however, of the power of the larvae to pass through the tissues of the host. Nor it is precisely determined by what route the larvae migrate from the intestine to the liver and from the liver to the lungs of the experimental animal. It is thought that there are three routes by

which the larvae may pass from the intestine to the liver: (1) by the blood vessels distributed on the wall of the alimentary canal; (2) by the common bile duct, and (3) by penetrating through the intestinal wall and through the surface of the liver. The route last mentioned seems most probable from Experiment XXXVI, but none of my attempts to determine this point were successful. Stewart supposed that the larvae penetrating the alimentary wall are carried into the liver by one of two routes, by the mesenteric venules or by the bile duct, he did not determine which.

Between liver and lungs two courses of migration are supposed to exist, the blood vessel and the body cavity. Experiments XXIII and XXVI seem to favor the former, but Experiment XXXVI suggests that the latter course is possible. Further investigations on this subject are being carried out.

Human Experiments with Ascaris Larvae

On August 8, 1917, the writer swallowed 35 larvae of various sizes taken from the lungs of a guinea-pig killed on the seventh and eighth days, respectively, after two feedings. Examinations of the feces for the eggs on September 1, 8, 13, 17, October 19 and November 8 were negative, probably because the larvae were not completely developed. Hence larger larvae from the lungs or trachea were used in the second experiment. October 19, 1917, five specimens (2 mm. long) were swallowed. These were taken from the lungs of the guinea-pig killed on the eighth day after feeding. On November 7, 50 specimens were ingested, 1.61 mm. long and coming from the trachea of a guinea-pig killed on the ninth day after feeding. Examination of feces for the eggs on December 8, 18, 29 and January 8 were negative. On January 21, 75 days after the last feeding, however, numerous eggs were present in the feces. Since that time it has often been possible to find the eggs in the feces, and since there is no other explanation of the infection it is reasonably certain that it is the result of these experiments.

Morphological Changes in the Larvae During Development

The detailed study of this part of the subject is not yet complete. The embryo in a mature egg coils its body once or twice within the shell and is actively mobile. The larva just emerged from the egg-shell is slender, cylindrical in shape, with a rounded anterior and pointed posterior extremity, the latter conical and slightly turned

toward the dorsal side. The anus is ventrally situated at the base of the conical portion of the posterior extremity. A small portion of each extremity is homogeneous and transparent in appearance, the anterior being a little larger than the posterior end. The greater portion of the body is internally granulated, indicating the intestine and the lower part of the esophagus.

The larvae in the intestine, liver, and lungs are also very active and are always coiled, like the adult form. It is perhaps a characteristic movement of the species. Embryos measured varied in size from 0.23 to 0.27 mm. in length and from 0.013 to 0.017 mm. in breadth.

The larvae in the liver as they mature become differentiated in organization. Three lip-like processes on the front of the body become more visible. The esophagus, which occupies one-third of the intestinal tract, takes a definite form, its posterior end being slightly swollen, its wall very thick, and the lumen very thin but clearly visible. Near the middle of the esophagus there is presently recognizable a group of cells which should be the foundation of the nervous ring of the adult form. The intestine is also differentiated in that the thick wall and the thin lumen are distinguishable. It runs straight along the median line of the body and occupies two-thirds of the length of the digestive canal. A cellular layer of the body wall may be easily distinguished from that of the intestinal wall by a thin space which is regarded as the body cavity. The genital area appears as a distinct cell in the ventral side at about one-third of the body length from the posterior end.

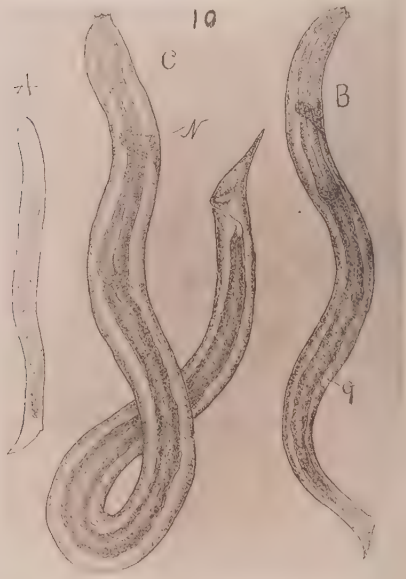
Table 1 shows the measurements of various parts of the body in living specimens.

TABLE 1.—Data from Living Specimens

	Total Length mm.	Maximum Breadth mm.	Length of Esophagus mm.	Distance of Anus from Posterior End mm.
1	0.364	0.021	0.1	0.25
2	0.417	0.02	0.117	0.026
3	0.435	0.025	0.107	0.022

In the lungs the larvae become thrice or four times as large as those in the liver, but there is no remarkable difference in structure. The differentiation of organs become daily more apparent. They vary in size according to the date of infection and are found in the blood vessels, alveoli and bronchi. The intestine of the fully grown larva in the lungs is yellow or light brown in color, perhaps due to food particles.

Table 2 shows the measurements of specimens mounted in potassium acetate or in glycerin.



del S. O. Yoshida.

TABLE 2.—Specimens Mounted in Potass. Acetate or Glycerine

	Body Length mm.	Maximum Breadth mm.	Length of Esophagus mm.	Distance of Anus from Posterior End mm.
1	1.90	0.056	0.28	0.062
2	1.70	0.052	0.26	0.061
3	1.62	0.053	0.26	0.06
4	1.45	0.051	0.25	0.06
5	1.23	0.047	0.23	0.055
6	1.014	0.042	0.2	0.053
7	1.857	0.042		
8	0.857	0.034	0.178	0.050
9	0.846	0.039	0.228	0.050
10	0.666	0.038		
11	0.657	0.030	0.214	0.052
12	0.571	0.028	0.125	0.035
13	0.385	0.028	0.128	0.035
14	0.357	0.028	0.072	0.032
15	0.300	0.021	0.064	0.025

The larvae in the trachea and alimentary canal are similar to the fully developed ones in the lungs.

It is my pleasant duty to express my sincere thanks to Prof. A. Sato, Director of the Osaka Medical Academy, for his kind advice and help during the course of this work. I am also indebted to several other persons who have given me cordial assistance in the investigation.

Explanation of Plate

Figs. 1-9. Stages of development of Ascaris egg. × 300.

Fig. 10. Embryo just hatched in the intestine of guinea-pig. × 233.

Fig. 11. Larva from the liver of guinea-pig. × 233.

Fig. 12. Larva from the lung of guinea-pig. × 233.

Fig. 13. Larva in the liver of guinea-pig. × 500.

Fig. 14. Larva in the lung of guinea-pig. × 300.

Abbreviations: g, genital cell; n, nerve ring; o, dust; v, alveola.

[Editor's Note.—This paper appeared originally in Japanese, in the *Tokyo Iji Shinshi* (Tokyo Medical Weekly Journal). Through the courtesy of the author and the cooperation of the editor, a copy of the original plate is included in each copy of this journal.]

ON A SPECIES OF HEDRURIS OCCURRING COMMONLY IN THE WESTERN NEWT, *NOTOPHTHALMUS TOROSUS* *

ASA C. CHANDLER

It has recently been pointed out by Ward and Magath (1916) that the nematode fauna of North American freshwater fishes is almost unknown. From a survey of the literature this state of affairs appears to be almost if not quite as true of the nematodes of other cold-blooded vertebrates in North America.

While making stomach examinations of the Western Newt or water-dog, *Notophthalmus torosus,* the writer found a large percent of specimens captured in the vicinity of Corvallis, Oregon, infested by a nematode which was evidently a species of Hedruris. The structure and ecology of this worm are so unique and proved so interesting and the references to it in the literature are either so slight or so difficult of access, that it seemed worth while to draw the attention of American helminthologists to it. Hitherto the members of the genus Hedruris have been looked upon as rare worms, and many helminthologists are no doubt unfamiliar with them.

According to Perrier (1871), whose account of *Hedruris armata* is by far the most thorough that can be found in literature of any species of Hedruris, these peculiar worms were first known to Rudolphi, but Perrier says: ". . . neanmoins leur caractéres si particuliers lui ont completement echappé, et il nous semble assez difficile de reconnaitre, comme le fait Schneider (Monogr. der Nemat., p. 107), *l'Hedruris androphora* dans *l'Ascaris acuminata* du savant helminthologiste." Nitzsch (1821) described the first known species, *H. androphora,* in a readily recognizable manner, and proposed its separation from the genus Ascaris into a new genus Hedruris. The first and only good description of *H. androphora,* the type species, is given by Schneider (1866). This species is found in various European amphibians and has been recorded from *Bufo calamita, Bombinator igneus, Triton cristatus, Lissotriton punctatus* and *Proteus anguineus,* always attached to the mucous membrane of the stomach.

Leidy (1851), apparently unaware of the existence of *H. androphora,* described a worm, evidently a Hedruris, which he found in the stomach and commencement of the small intestine of *Clemmys guttata,* and named it *Synplecta pendula.* Baird (1858) very inadequately

* Contribution from the Zoological Laboratory, Oregon Agricultural College.

described a Hedruris from the axolotl, *"Siredon mexicanus";* the larval form of *Ambystoma tigrinum,* giving it the name *Hedruris siredonis.* Perrier (1871) described another species, *H. armata,* from the back part of the mouth cavity of *Chrysemys picta.* His description is, as already remarked, by far the most thorough of any which can be found in the literature. From the time of Perrier's paper to the present, no further research on this unique genus has been done, and very few references to it, even of the most casual kind, can be found.

The following is a diagnosis of the genus based on the descriptions of other authors and on the writer's observations on the species found in *Notophthalmus torosus:*

Cylindrical whitish worms, tapering from a fairly robust posterior end to a slender anterior end; surface of body more or less finely striated; mouth provided with two pairs of lips, a median and a lateral (Figs. 2 and 3); median lips (Fig. 2) thin, chitinized, concave on the upper surface, and attached to the worm only by the middle portion of the base; lateral lips (Fig. 3) enclosed by median ones, thick with elaborate chitinous skeleton, and united by a chitinous commissure; esophagus long, slender, muscular, crowned by a festooned chitinous ring and penetrating valve-like into the intestine, its lumen four-cornered, not three-cornered; intestine straight, cylindrical, terminating in a chitinous rectum in the female.

Female attaching itself to mucous membrane of host by means of posterior portion of body, which can be invaginated sucker-like and has attached to it a chitinized hook resembling the claw of a cat, the latter being used to hook into the mucous membrane of the host and to draw it up into the interior of the invaginated portion of the body (Fig. 5); female reproductive system double, with dilations of the oviducts which act as seminal vesicles (Fig. 7); vulva posterior, shortly anterior to the anus; eggs (Figs. 8 and 9) oval, provided with terminal opercula as in Trichuridae, and containing developed embryos when deposited.

Male smaller and slenderer than female, always rolled about his mate by about three spiral turns of the body; surface of body, anterior to anus, in contact with female on inner surface of spiral coil is provided with 15 to 20 rows of quadrangular tubercles, reminding one forcibly, both in appearance and function, of the elevations on certain kinds of non-skid tires (Fig. 6); spicules short, equal, crescent-shaped, with or without a small gubernaculum; one preanal and at least six postanal papillae.

Habitat of species so far known always stomach or back of mouth cavity of amphibians and pond turtles.

In many of the respects named above, Hedruris is absolutely unique among nematodes, and it is evidently a very highly specialized form. There can be no doubt, however, that it finds its nearest relatives among the Spiruroidea, and should be considered representative of an aberrant family belonging to this group as suggested by Railliet (1916). Perhaps the forms which approach nearest to it are the species of the genus Habronema (cf. figures by Ransom, 1913). The form and arrangement of the lips and the armature of the mouth; the chitinous crown for the esophagus (Figs. 2 and 3) and the general

form of this organ; the form of the vagina, with a muscular mass surrounding its distal end (Fig. 5); the dorsal curvature of the tail of the female (Figs. 1 and 5); the chitinized rectum; the form, but not arrangement, of the male caudal papillae; are all strikingly similar to conditions found in Habronema. The cervical papillae of Habronema are also remarkably suggestive of the tactile spines described and figured by Perrier for *Hedruris armata*. Furthermore, the ventral surface of the body of the male of Habronema, anterior to the anus, is remarkably like that in Hedruris. Similar modifications occur in Arduenna and other spiruroids. On the other hand, Hedruris is less specialized than other spiruroids in the short, equal spicules, and more specialized in the development of the caudal adhering apparatus of the female, in the presence of distinct seminal vesicles, in the relation of the sexes, and also in the degree of specialization of the lips.

As already shown, four species of Hedruris have already been described, and considerable confusion exists as to the status and relationships of the species. In an endeavor to ascertain the identity of the species found by the writer in *Notophthalmus torosus* a careful comparison of the published descriptions of the species has been made and certain conclusions have been reached, though the incompleteness of the descriptions makes it necessary to assume more than is desirable.

The first described species, and therefore the type of the genus, is *H. androphora* Nitzsch. It may be defined as a small species, not exceeding 10 mm. in length in the female and 8 mm. in the male, within distinctly striated cuticula, and with mammillated eggs; it occurs as a stomach parasite of various European amphibians. The only other well-described species is that of Perrier, *H. armata,* which differs from *H. androphora* in its larger size (♀ 23 mm., ♂ 20 mm. long), in its distinctly striated cuticula in both sexes, in its non-mammillated eggs, and in the presence of tactile cervical spines; it occurs as a pharyngeal parasite of the American *Chrysemys picta,* having been obtained from a specimen in the menagerie of the Paris Museum of Natural History.

Perrier, when describing his species, was evidently unaware of the species described by Leidy twenty years before under the name *Synplecta pendula.* So far as Leidy's description goes, it appears to harmonize perfectly with that of Perrier. The size of Leidy's specimens (12.5 mm. to 23 mm. for the female and 8 to 10 mm. for the male), the marked striation of the body, the number of caudal papillae of the male, and the lack of any mention of the eggs being mammillated on the sides, all ally this species with *H. armata* and not with *H. androphora.* Leidy does not mention cervical spines in his species, but one would hardly look for such a minute detail in such a superficial description. Leidy's specimens were taken from the stomach and commence-

ment of the small intestine of *Clemmys guttata,* which, like *Chrysemys picta,* is a common pond turtle in Eastern United States. It appears very likely, therefore, that *H. armata* and *H. pendula* are synonyms, in which case the latter name would have to stand. Stiles and Hassall (1894) refer to specimens of Hedruris in Leidy's collection in the University of Pennsylvania taken from *"Ambystoma mexicanum"* and *"Nanemys guttata"* as *H. androphora.* Baird's *H. siredonis* is so meagerly described that its identity can only be inferred from circumstantial evidence. His description is based on a single female specimen, probably immature, taken from an axolotl from Mexico. The size, 13 mm., and marked striation of the body, the only two characteristics of specific value mentioned, are evidence that it is not *H. androphora.* There is nothing except the host to distinguish it from *H. armata* or *H. pendula,* and it would naturally be included with them were it not for the fact that the specimens found by the writer in the western newt evidently represent a species distinct from either *androphora* or armata, and the probability is in favor of Baird's specimen being identical with the writer's species. So far as it goes, Baird's description agrees with the species found in *Notophthalmus torosus,* and its geographic occurrence and host both point to a probable identity of the two worms. Provisionally, therefore, the writer will refer to his specimens as *H. siredonis* Baird.

As found in *Notophthalmus torosus,* the last species appears to be more or less intermediate between *H. androphora* and *H. armata.* It is intermediate in size, the full grown females (Fig. 1) being 16 mm. to 17 mm. long, with a maximum width of from 0.5 mm. to 0.6 mm., while the males are 8 mm. to 10 mm. long by 0.23 mm. wide. The body of the female is very coarsely striated; the striations are about 50μ apart, and each in turn is marked by from 12 to 15 exceedingly fine striations (Fig. 5). The body of the male, on the other hand, is very indistinctly striated, the striations being so light that when the body is curved they cannot be seen at all on the greater curvature. Male specimens cleared in carbolic acid show no evidence whatever of striation, whereas even the fine substriations of the female can be observed clearly. Often the striation of the male seems to be entirely missing, at least on parts of the body. This indistinctness of striation in the male is in contrast with the condition in *H. armata,* in which Perrier says the male is striated evenly from head to tail. There is no evidence whatever of cervical spines in *H. siredonis.* The male possesses a small gubernaculum in addition to the spicules, and has altogether ten pairs of postanal papillae, seven near the midventral line, two placed more laterally near the top of the tail, and one just behind the anus (Fig. 6). The eggs (Figs. 8 and 9) resemble those of

H. androphora in their operculation, and also in the mammillated surface, a feature which is conspicuously absent from *H. armata.* The lips (Figs. 2 and 3), both median and lateral, are shaped a little differently in *H. siredonis* than in *H. armata.*

In other characteristics of structure and anatomy *H. siredonis* agrees with Perrier's description of *H. armata* (Fig. 7). The reproductive system is built on the same plan, but the ovaries with their radially arranged eggs are longer, and the seminal vesicles are more distinctly marked off. The branches of the uterus also are larger and contain a greater number of eggs than is the case with *H. armata,* according to Perrier's figure. Hedruris is the only nematode known to the writer in which a completely distinct seminal vesicle is present. Hall describes a slight dilation of the anterior ends of the uteri of *Dermatoxys veligera* as a seminal vesicle; in this genus there is also a slender oviduct connecting ovary and uterus.

To sum up, the species of Hedruris may be characterized as follows:

1. Size small, ♀ not exceeding 10 mm. in length; indistinctly striated if at all; eggs mammillated; in stomach of European amphibians.
H. androphora Nitzsch.

2. Size medium, ♀ 16 mm. to 17 mm. long, distinctly striated; ♂ indistinctly or not striated; eggs mammillated; in stomach of *Notophthalmus torosus* and axolotl. *H. siredonis* Baird.

3. Size large, ♀ 23 mm. long; both sexes distinctly striated; a pair of cervical tactile spines; eggs not mammillated; in pharynx of *Chrysemys picta* (probably identical with *H. pendula* Leidy from stomach of *Clemmys guttata*).
H. armata Perrier.

Unlike any previously described Hedruris, *H. siredonis* is an abundant parasite, at least in the vicinity of Corvallis. The majority of all specimens of *Notophthalmus torosus* examined are infested, frequently by a few worms, but sometimes by 20 to 25 pairs. Very often the females exceed the males in number, i. e., there are frequently females unaccompanied by males. Unattached males have never been found. Since it is always adult ripe females which are unaccompanied, it is to be presumed that the males die sooner than the females, and pass out of the digestive tract of the host. Nitzsch, however, states that he has found *young* females of *H. androphora* unaccompanied, assuming that they were too delicate to support the males.

That the worms when present in large numbers have an injurious effect on their hosts is evident. The mucous membrane of the stomach around the places where the worms adhere is often considerably swollen, and the point of attachment of the worms can sometimes be seen from the outer surface of the stomach. Perrier demonstrated

that the caudal claw of the female worm is perforated and connected with a gland, thus imitating closely the fang of a solenoglyph snake, and, as suggested by Perrier, the probable function of the secretion of the gland is to irritate the tissues of the host sufficiently to cause a swelling, the latter making the attachment of the worm the more secure. On several occasions the writer has observed that heavily infested newts have been undersized and thin, and contain little or no food in the stomach.

H. siredonis, as observed in life, is an extremely interesting animal. The invaginated posterior end of the female is seldom entirely everted, usually only far enough to throw the claw into such a position that it can seize a surface in contact with it and draw it back into the invagination. The worms are in general sluggish in movement, and appear to be incapable of the rapid swimming in which many nematodes indulge. The male worms, when slid off from the females, can only partially uncoil, and the permanent nature of the coils is demonstrated by the fact that when freed males are immersed in hot formalin the posterior end invariably coils in such a way as to leave a tube corresponding to the diameter of the female worm.

The life history of Hedruris is as yet unknown, outside of the fact, stated by Perrier, that the embryos escape from the eggs upon slight pressure, or sometimes spontaneously when in fresh water, probably due to osmosis. By analogy with other spiruroid worms, and from the fact that very small individuals are never found in the newts, it is probable that the eggs or embryos are swallowed by an intermediate host in which the early stages of development are passed, and that infection of the definitive host is brought about by feeding on the intermediate host.

Literature Cited

Baird, W. 1858.—Descriptions of Five New Species of Entozoa. Proc. Zool. London, pt. 26, 224-225, pl. 52.

Leidy, J. 1851.—Contributions to Helminthology. Proc. Acad. Nat. Sci., Phila., 5:239-240.

Nitzsch, C. L. 1821.—Ascaris, in Ersch and Gruber's Encyclopedia, 6:44-49, 2 pl.

Perrier, Ed. 1871.—Recherches sur l'organization d'un nematoide nouveau du genre *Hedruris.* Nouv. arch. du mus. d'hist. nat. de Paris, 7:1-60, 2 pl. [Abstract of same under title "Sur l'organization d'un espece nouvelle de nematoide appartenant au genre *Hedruris,*" Compt. rend. acad. des sci., Paris, 72 (12), 337-339.]

Railliet, A. 1916.—La Famille des Thelaziidae. J. Paras., 2:99-105.

Ransom B. H. 1913.—The Life History of *Habronema muscae* (Carter) a Parasite of the Horse Transmitted by the House Fly. U. S. Bur. An. Ind., Bull. 163, 36 pp., 41 figs.

Schneider. 1866.—Monographie der Nematoden. Berlin, p. 107-108, pl. IV, fig. 8.

Stiles, C. W., and Hassall, A. 1894.—A Preliminary Catalogue of the Parasites Contained in the Collections of the U. S. Bur. An. Ind., U. S. Army Med. Mus., Biol. Dept. U. of Penn. (Coll. Leidy) and in Coll. Stiles and Coll. Hassall, Vet. Mag., 1: 341.

Ward, H. B., and Magath, T. B. 1916.—Notes on some Nematodes from Freshwater Fishes. J. Paras., 3: 57-64, 1 pl.

Explanation of Plate

Fig. 1.—Adult female *Hedruris siredonis*. × 11.

Fig. 2.—Anterior end of body of ♂ *H. siredonis* showing median lip. Note chitinous commissure connecting lateral lips, and chitinous crown of oesophagus. × 132.

Fig. 3.—Anterior end of young ♀, showing lateral lip. Note chitinous crown of esophagus; comparison of Figures 2 and 3 will show that the lumen of the oesophagus is four-cornered. Note also excretory pore and nerve ring. × 110.

Fig. 4.—Male and female *H. siredonis* in situ. × 11.

Fig. 5.—Posterior end of adult ♀ *H. siredonis*, showing posterior portions of digestive and reproductive systems, and caudal adhering apparatus. × 28.

Fig. 6.—Posterior end of ♂ *H. siredonis*. Note longitudinal rows of quadrangular tubercles, the crescent-shaped spicules, the spearhead-like gubernaculum, and ten pairs of post anal papillae.

Fig. 7.—Female reproductive system of *Hedruris armata*. Note arrangement of eggs or ovaries and form and position of seminal vesicles. After Perrier.

Figs. 8 and 9.—Eggs of *H. siredonis*. × 260.

Abbreviations Used on Plate

a — Anus
au — Ascending uterus
bu — Branches of uterus
dl — Dorsal lip
du — Descending uterus
ep — Excretory pore
int — Intestine
ll — Lateral lip
lo — Lumen of oesophagus
nr — Nerve ring
oes — Œsophagus
on — Œsophageal nerve
ov — Ovary
ovd — Oviduct
ph — Posterior hook
ps — Posterior sucker
psv — Peduncle of seminal vesicle
sv — Seminal vesicle
u — Uterus
up — Uterus proper
va — Vagina
vu — Vulva
wo — Wall of oesophagus

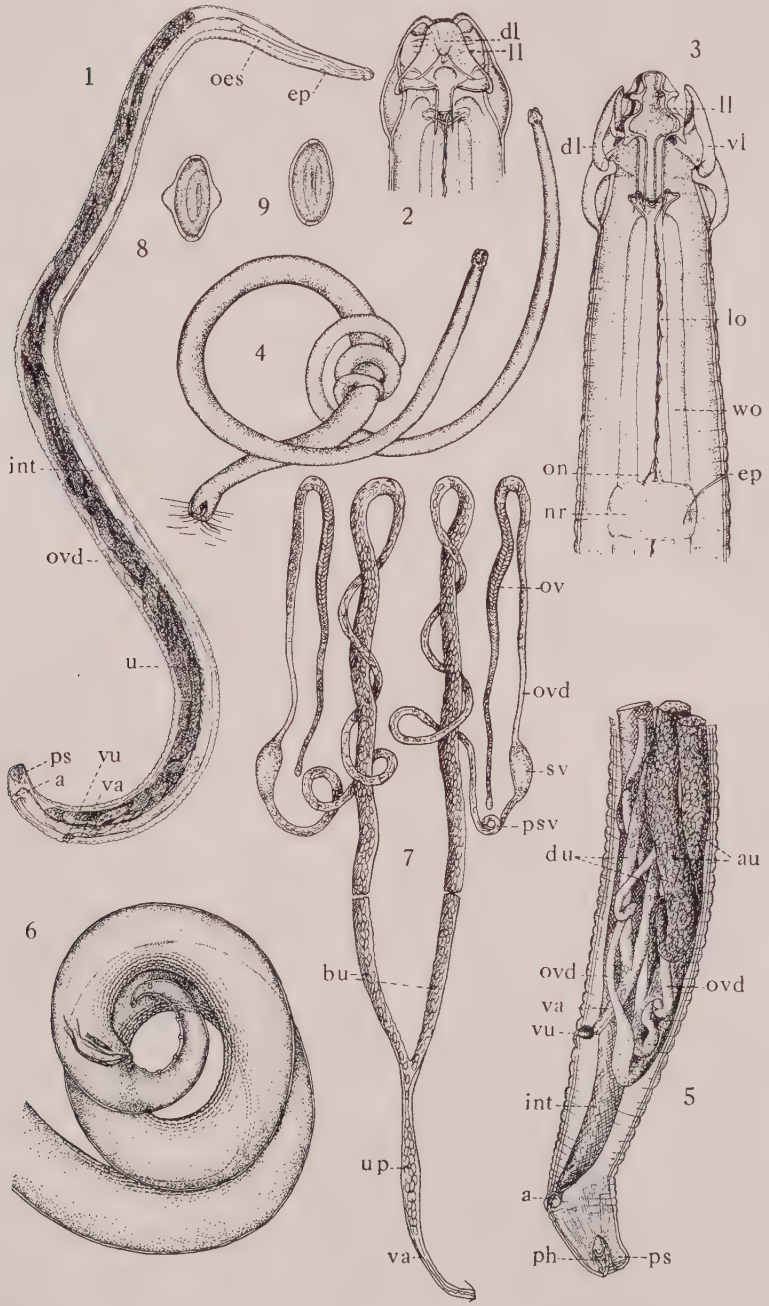

1
oes
ep
int
ovd
u
ps
a
vu
va
2
dl
ll
3
ll
dl
vl
lo
wo
on
ep
nr
4
5
ovd
ovd
va
vu
int
a
ph
ps
6
7
bu
up
va
ov
ovd
sv
psv
du
au
8
9

PLATE IX

OBSERVATIONS ON *MICROPHALLUS OVATUS* SP. NOV. FROM THE CRAYFISH AND BLACK BASS OF LAKE CHAUTAUQUA, N. Y.*

H. L. Osborn

The recent account by Yoshida (1916: 76-82) of an unnamed trematode infesting the liver, ovary and other tissues of Japanese crustacean, living in burrows between tides on the seashore near Osaka, recalls some observations which I made several years ago, but which have never as yet been published. Though Yoshida did not designate the form which he described, his account and drawings show it to be a member of the genus Microphallus (Ward, 1901: 184). For convenience, I will refer to it as *M. japonicus,* which name I would propose to call it.

Especial interest attaches to the case of *M. japonicus* as another instance of that remarkable correspondence between parasite and host organ infected which has been noticed by various writers and in particular by Johnston (1913: 272) in Australian frogs. *M. japonicus* is the third case now known where the liver or adjoining thoracic viscera of the crayfish or some other crustacean is infected by a species of Microphallus.

I first noticed the Chautauqua species in the crayfish in the summer of 1898 and saw its close relationship with *M. opacus* of Ward (1894: 173 and 1901: 175). I found it later also in the stomach of the black bass and of the bullhead from Lake Chautauqua. In the liver of the crayfish (Fig. 1) it forms large whitish spherical cysts among the tubular cecae of the infected organ. In August, 1902, it was found in every individual of a group of twenty-five examined. In one instance 22 cysts were found on one side and 18 on the other, totalling 40 cysts for the organ. One case, very badly infected, showed about 100 cysts. In the stomach of the black bass partly digested crayfish are often found, and specimens of the worm in various stages are seen which are thus traced to the crayfish as the source of infection. The bass is more often infected than the bullhead, the diet of the latter regularly being more largely molluscan. The Chautauquan form shows marked resemblances to *M. opacus,* but there are certain differences which seem at present to justify distinguishing them and I am proposing to give it the name *M. ovatus* in allusion to its shape.

---

*Contributions from the Biological Laboratory of Hamline University, St. Paul, Minn.

M. ovatus differs from *M. opacus* in its habits, the latter infecting not the liver but the genital glands and the space above the cephalothorax. The Japanese species infects the liver like *M. ovatus,* and also the ovary like *M. opacus.* The final host of *M. opacus* also is different. Ward examined *Micropterus dolomieu,* "yet none were infected"; though it was found in the yellow perch, but the chief host is *Amia calva,* where hundreds were found in the intestine (vs. stomach in *M. ovatus*) just above the spiral valve.

The cysts of *M. ovatus* as removed from the liver are slightly elongate, measuring 1 mm. by 0.7, slightly smaller than Ward's figures, 1.28 mm. by 0.9, and larger than *M. japonicus,* which averages 0.521 mm. by 0.418. The encysted worm is bent double ventrally; under slight compression enough of its structure is visible to show that it is nearly or completely developed, as shown in Figure 2, drawn from a specimen immediately after its escape from the cyst. Inside the cyst the anterior end of the body can be seen actively moving. The crustacean host is thus evidently an intermediate host interpolated between the primary host and the definitive one, as happens in the case of Clinostomum and various other genera of trematodes.

Specimens of the free mature worm from the bass (Fig. 3) had the body densely packed posteriorly with the genital glands, but clear anteriorly so that this part was very mobile, pushing itself forward and seeming to adhere slightly by means of the small oral sucker, while a wave of contraction swept backward the whole length of the body giving it a dumb-bell shape as it passed the center and passed off over the hind end without influencing its form. *M. ovatus* is larger than *M. opacus* and presumably than *M. japonicus,* the adult of which is not known. The dimensions of the body in a mature specimen from the bass were 2.6 by 1.7 mm. Another worm drawn under slight compression measured 3.7 by 1.2 mm. One from the bullhead gave 3 by 1.8 mm. Ward gives 1.7 by 1 mm. for *M. opacus,* and *M. japonicus* from the crustacean cysts in maximum specimens was 0.833 by 0.325 mm. As the organization of the encysted specimens of *M. japonicus* is almost completely developed, one must conclude that the mature worms are much smaller than the American forms. The cuticula is spinous throughout as in the Japanese form, but unlike the *M. opacus* according to Ward, who says (1894) that the entire surface is free from spines.

The terminal oral sucker measured 0.125 mm. in a section of the worm, in another case drawn from a living specimen it measured 0.1 mm. Ward gives 0.155 in *M. opacus,* and Yoshida 0.05 mm. In the size of the ventral sucker, *M. ovatus* also differs from the others,

measuring 0.16 and 0.175 mm. in two cases, as against 0.210 in *M. opacus* and 0.035 in *M. japonicus.* The suckers are relatively larger in *M. opacus* as the bodies are smaller. In *M. ovatus* the ventral sucker is located on the level of the fifth eighth of the body length. It is located at about the same point in *M. opacus,* but in *M. japonicus* it is more posterior, being placed at the sixth eighth of the total length. The genital opening is located on the level of the center of the ventral sucker and in immediate contact with its left side. The excretory opening is placed at the extreme posterior end of the body.

The digestive apparatus is very poorly developed, the least of any of the members of this family. The very feebly developed pharynx, slightly more remote from the oral sucker than is that of *M. opacus,* has a length of less than 0.1 mm. and a diameter of only 0.05 mm. A long, slender esophagus leads back to a small triangular sack which can hardly be said to be forked, thus contrasting with the distinctly developed ceca of *M. opacus* and the still more elongate ones of *M. japonicus.* Sections and total preparations show a feebly developed epithelium in the interior of this digestive apparatus, but no conspicuous glands connected with esophagus.

The dorsally located excretory bladder is V-shaped, each side is broad and conspicuous, measuring about 0.4 mm. in length and having a diameter of about 0.1 mm. In specimens from the crayfish the cavities of these vesicles are filled with highly refractive globules doubtless the stored excretions resulting from the metabolisms of the encysted animal, similar to those found in various other cases of encysted stages (e. g. Faust, 1917: 42), but not found in the mature worms from fishes.

Possibly the drawings of Yoshida (Fig. 2) may be interpreted to mean the same thing. The excretory vesicles of living specimens were watched on several different occasions, but no pulsations or other movements were detected. A slender tube was traced forward from each horn of the bladder to a point on the level of the pharynx. These lateral vessels gave rise to smaller vessels which in turn gave rise to capillaries and terminated in flame cells. As in *M. opacus* and *M. japonicus* the cerebral ganglion and the lateral nerve cords are very noticeable even in preserved preparations.

All of the reproductive organs are confined to the posterior half of the body. The compact globular testes lie nearly opposite each other, they are distinct and not in contact with the excretory vesicles, whereas in *M. japonicus* they are apparently in contact. Their ducts join and form a common duct which, passing anteriorly to the ventral sucker, enters the posterior side of the large seminal vesicle which

lies directly in front of the ventral sucker. The organ is also conspicuous in *M. japonicus,* where it is called by Yoshida "the semilunar organ." In mature specimens it was filled with living spermatozoa. It tapers distally (see Fig. 5) to form a passage leading directly to the eversible penis and surrounded by cells of the prostate glands. As in all the Microphallinae, these parts are not enclosed in a cirrus sack after the manner of many trematodes. There is no atrial pocket common to the penis and metraterm as in Levinseniella (Lühe, 1899; 124), but these organs reach the surface entirely separately, as shown in Figure 4. The globular ovary, larger than the testes, lies on the level of the ventral sucker and on the right side of the body. Its duct meets the vitelline ducts close behind the ventral sucker as in *M. opacus,* there is no seminal receptacle, the uterus in mature stages is much enlarged and filled with eggs, in encysted stages from the crayfish its empty windings can be seen. The course of the uterus in a mature specimen is shown in Figure 3. After leaving the yolk receptacle it runs to the posterior end of the body, then bending, runs forward and back again on that side externally to and partly enclosing the vitellaria it then crosses to the opposite side and again runs forward and back in close relation with the vitellarium; finally it runs directly forward to the genital opening. The uterus contains great numbers of small dark-shelled operculated ova which measure 25 by 12μ, slightly smaller than those of *M. opacus,* which, according to Ward, measure 0.03 to 0.04 by 0.015 to 0.02 mm.

The vitellaria are more compact apparently than in *M. japonicus,* where the follicles are shown as if quite distinct and apparently are less deeply lobed than in *M. opacus.* They are located externally to the spermaries and extend beyond them reaching as far anterior as the level of the ventral sucker. Their position is intermediate between that of the *M. opacus* and *M. japonicus,* where they are wholly behind the spermaries or wholly anterior to them, respectively.

The habits of *M. ovatus* are slightly different from those of *M. opacus.* From the accounts of Ward one would consider that the liver of the crayfish is not infected, as the infection is said to be seated in the space above the cephalothorax and sexual organs. The final host also is different as shown by Ward who examined *Micropterus dolomieu,* "yet none were infected," though it was found in the yellow perch, but it is found in *Amia calva* where hundreds were found in the intestine just above the spiral valve. We note also that the Japanese species inhabit the liver of its crustacean host like *M. ovatus* as well as the "ovary and hypodermis" like *M. opaca.*

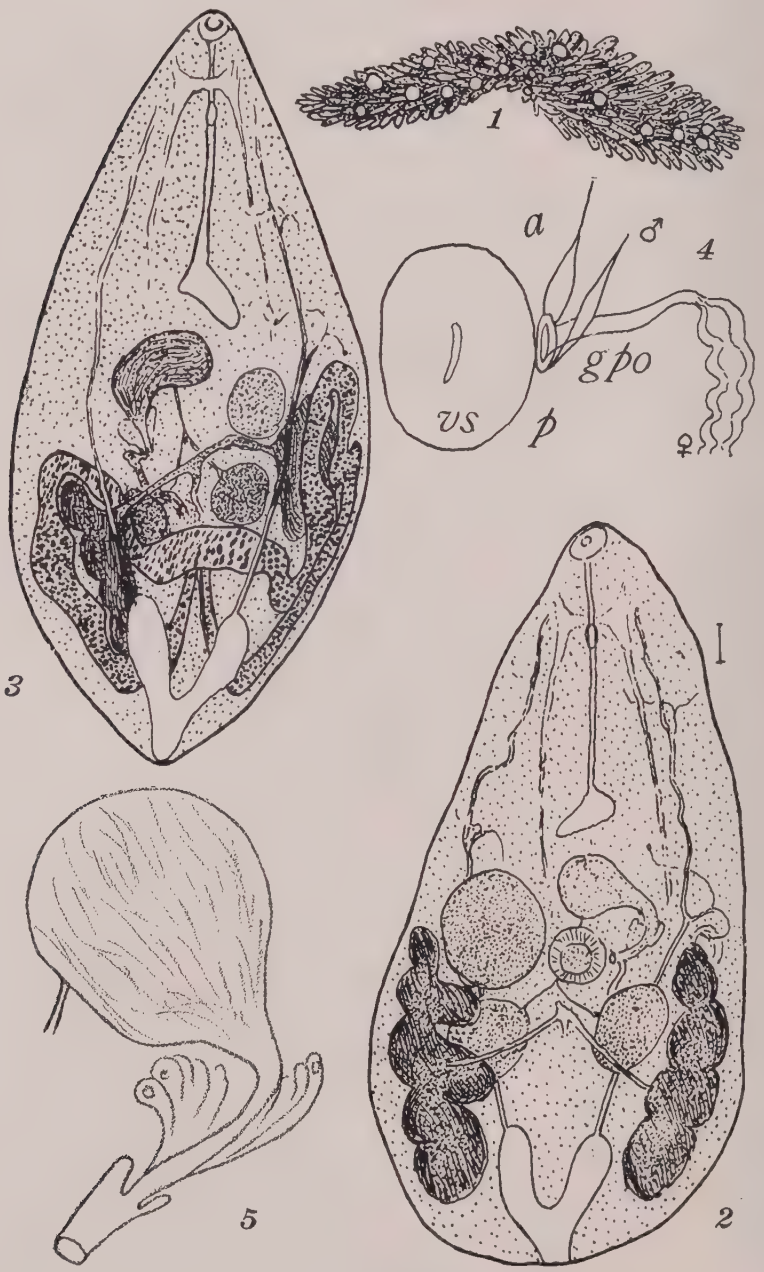


PLATE X

The differences noted between these three species are shown briefly in the following table.

Microphallus	*Opacus* Ward	*Japonicus* Yoshida	*Ovatus* Osborn
Infect as larva	Genital glands, cephalothorax of Cambarus	Liver, ovary and cephalothorax of Helice	Liver, of Cambarus
Adult	Intestine of Amia	.....	Stomach of bass
Maximum length	1.7 mm.	0.85 mm.	3.7 mm.
Cuticula	Non-spinous	Spinous	Spinous
Ventral sucker	0.210 mm.	.....	0.175
Intestinal ceca	Distinctly formed	Moderately long	Very rudimentary
Vitellaria	Deeply lobed, wholly posterior to testes	Wholly divided; wholly anterior	Slightly lobed, enveloping testes

References Cited

Faust, E. C. 1917.—Life History Studies on Montana Trematodes. Illinois Biol. Monog., 4: 1-121.

Johnston, S. J. 1913.—Trematode Parasites and the Relationships and Distribution of their Hosts. Rep. Australian Assn. Adv. Sci., Melbourne Meeting, 14: 272-278.

Lühe, M. 1909.—Parasitsche Plattwürmer, I. Trematodes, 126 pp.

Osborn, H. L. 1902.—Notes on the Trematodes of Lake Chautauqua. Sci., n. s., 15: 573-4.

Ward, H. B. 1894.—On the Parasites of the Lake Fish; Notes on *Distomum opacum* n. sp. Proc. Am. Mic. Soc., 15: 173-182.

1901.—On the Structure of the Copulatory Organs of Microphallus nov. gen. Trans. Am. Mic. Soc., 22: 175-187.

Ward, H. B., and Whipple, G. C. 1918.—Fresh-Water Biology, Wiley and Sons, N. Y., 401.

Yoshida, S. 1918.—On a Trematode Larva Encysted in a Crab. Jour. Parasitol., 3: 76-79.

Exlanation of Plate

Fig. 1.—View of the liver of Cambarus from Lake Chautauqua, N. Y., showing the cysts of *M. ovatus* in place among the tubules of the organ.

Fig. 2.—Ventral view of immature specimen of *M. ovatus* immediately after its escape from the cyst, based on camera lucida drawings from compressed living specimens and total preparations. Scale equals 0.1 mm.

Fig. 3.—Dorsal view of mature specimen, taken from the stomach of *Micropterus dolomieu*, showing the location of the coils of the uterus well filled with embryos, from free hand drawings.

Fig. 4.—View showing the relation of the ventral sucker, genital opening, penis and metraterm; drawn from living compressed specimen.

Fig. 5.—View from living specimen showing form of the seminal vesicle, prostate glands and eversible penis.

A NEW CYSTOCERCOUS CERCARIA*

H. S. Pratt

The cercaria described in this paper was found in the liver of *Goniobasis livescens* taken in the Oneida River near the outlet of Lake Oneida, in the State of New York. To it may be given the name *Cercaria fusca.* Twenty-three snails of the above mentioned species were examined, and of these, four were infected with small numbers of this cercaria. The color of the living worms is brown, the tail being much darker brown than the body. They were very sluggish when taken from the snails and moved very little, and made no swimming motions. The length of the extended live cercaria was about 3 mm., the body being about 1 mm. in length and the tail 2 mm.; the length of the flat tail-forks was about 0.5 mm. (Fig. 1a). Sporocysts were also found in the snails, which contained each from one to four or five cercaria in various stages of growth. The sporocysts are mostly lenticular in shape and 2 or 3 mm. in length. The largest of them contained but a single cercaria, which thus had the appearance of being encysted. One sporocyst observed, in which were several cercariae of different sizes, contained one apparently full grown, the tail of which projected freely from the sporocyst, giving the impression of a sporocyst with a tail.

These observations seem to support the statement of Faust (1918: 149) that the cystocercous cercariae may be cannibalistic. The largest cercaria in a sporocyst seems to feed upon, or at least absorb, the smaller ones until it is finally the only one left. I did not observe in the material studied any instance of the direct attack of a large cercaria upon a smaller one.

Cercaria fusca is similar to *C. brookoveri* Faust, 1918, in that the young distome is not surrounded by the walls of the anterior portion of the tail, as is the case in other cystocercous cercariae observed, but is joined with the tail by a short fold.

The shape of the body of the full-grown cercaria is pyriform (Fig. 2), the hinder end being the broader, where the width is 0.65 mm. The cross section is oval in shape, the thickness being 0.40 mm. The suckers are large. The oral sucker is an elongated organ 0.52 mm. long, 0.35 mm. wide and ventral in position. The acetabulum is

* This study has been made as a part of the ecological survey of Oneida Lake, being made under the direction of Prof. C. C. Adams of the New York State College of Forestry, Syracuse, N. Y.

just behind the middle of the body and measures 0.24 mm. in diameter. The pharynx has a diameter of 0.13 mm. and opens into a very short esophagus, which in turn leads into the two limbs of the intestine, each of which passes in the lateral area of the body to its hinder end. They are filled with a translucent secretion.

The median excretory trunk is short, passing from the hinder end of the body to a point immediately back of the ovary and dorsal to the testes, from which the two lateral limbs pass forwards in the dorsal area of the body to the right and left of the acetabulum. The testes and ovary are all spherical organs forming a group immediately back of the acetabulum and near the hinder end of the body. The two testes are close together in the same transverse plane, and in the ventral area of the body; each has a diameter of 0.13 mm. A large cirrus sac lies dorsal to the acetabulum. The ovary is slightly smaller than either of the testes and lies between and in front of them towards the dorsal

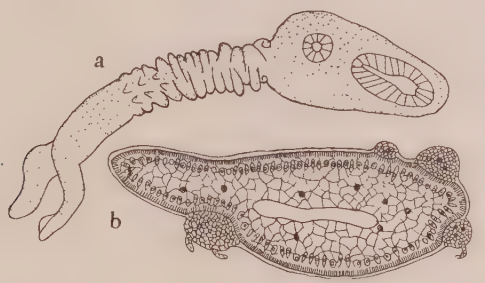


Fig. 1.—(*a*) Cercaria fusca, × 42. (*b*) Cross section of anterior portion of tail, × 400.

side of the body. A large receptaculum seminis lies alongside of the ovary. The uterus passes forwards to the oral sucker and thence back to the genital pore, immediately in front of the acetabulum. It contains a few large eggs which average about 78 by 49μ in size. The full grown cercariae are thus sexually mature and in this respect resemble *C. macrostoma* Faust.

The vitelline glands occupy the two lateral areas of the body from the oral sucker to the hinder end of the body, and consist of a large number of small follicles.

The tail of the full grown cercaria is flattened in shape and has a width of 0.27 mm. and a length of 2 mm. Each of the two forks of the tail is 0.5 mm. long and 0.27 mm. wide at its base. The tail is made up of two very distinct regions, the anterior two-thirds and the posterior third. The former region is distinguished by the presence of conspicuous warts and transverse ridges on its surface, similar to

the warts which have been observed in other cystocercous cercariae. It is also considerably thicker than the posterior region, having an oval cross section (Fig. 1, b). In the anterior portion is a large open space in the parenchyma which represents the excretory trunk.

The parenchyma of the tail is large meshed with numerous nuclei, and contains very conspicuous subcuticular cells and longitudinal and circular muscle fibers. The longitudinal fibers are wide bands lying close together and extending the length of the tail; it is by the contraction of them that the animal exercises the energetic swimming motions observed by Ward in *C. anchoroides* (1916: 12). The subcuticular cells form a very regular row which lies just within the longitudinal muscle fibers. They are mostly elongate or pear-shaped

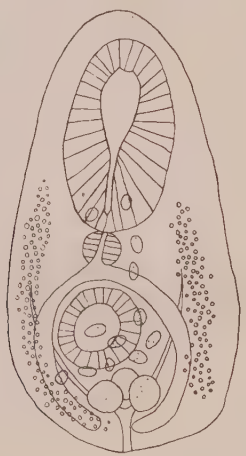


Fig. 2. Ventral aspect of distome with internal organs, × 125.

cells, the pointed ends of which extend towards or between the longitudinal muscle fibers. The warts contain no subcuticular cells and no muscle fibers but are composed entirely of a very close-meshed parenchyma in which lie a few nuclei, bounded on the outer surface by a cuticula. On the summit of many of the larger warts are one or two short, curved, finger-like projections. The circular and longitudinal muscle fibers and the subcuticular cells pass across the base of the warts, which thus lie entirely outside of them.

The posterior third of the tail together with the two flat tail-forks are similar in structure to the anterior two-thirds, except that they are much flatter and lack the warts. They also lack the wide excretory space of the anterior portion. Instead of this a narrow excretory canal passes through the middle, and branching at their base extends to the end of each of the forks.

Cercaria fusca is similar in structure to *C. macrostoma* Faust, and like that worm probably belongs to a species of distome allied to Allocreadium. It is also probable that *C. macrostoma* escaped from *Goniobasis pulchella,* inasmuch as that snail was present in the aquarium in which the worm was found swimming (Faust, 1918: 150).

The very regular arrangement of the subcuticular cells in the tail of *C. fusca,* as above described, and their frequent elongation between the longitudinal muscle fibers towards the cuticula would seem to give weight to the theory, so popular at present among helminthologists and morphologists generally (see Pratt, 1909), which regards them as a modified hypodermis, the function of which is to secrete the cuticula. The structure of the warts, however, does not give support to this theory, but disproves it, in that they contain no subcuticular cells but yet possess a well-defined cuticula. The subcuticular cells which pass along the base of a wart are separated from it by both the longitudinal and circular muscle fibers and bear no relation to it. They are portions of the parenchyma of central portion of the tail.

Literature Cited

Faust, E. C. 1918.—Two New Cystocercous Cercariae from North America. Jour. Parasit., 4: 148-153.

Pratt, H. S. 1909.—The Cuticula and Subcuticula of Trematodes and Cestodes. Am. Nat., 43: 705-729.

Ward, H. B. 1916.—Notes on Two Free-Living Trematodes from North America. Jour. Parasit., 3: 10-20.

OBSERVATIONS UPON *TRICHOMONAS INTESTINALIS IN VITRO*

MARK F. BOYD

Laboratory of Bacteriology and Preventive Medicine, Medical Department, University of Texas, Galveston

In a recent communication to this Journal I reported a successful cultivation of a strain of *Trichomonas intestinalis*. It is the purpose of the present communication to record some further observations upon this intestinal flagellate.

Fecal suspensions for subculturing the strain have been continuously secured from the same individual, but I have found that certain specimens of feces are unsuitable for the purpose of making suspensions, since in these the organisms will not grow. These unsatisfactory suspensions are characterized by an acid reaction to litmus and an intense bile staining of the saline above the fecal sediment. On the other hand, the suspensions in which good growth has been secured have been neutral to litmus and the supernatant saline has been colorless or only tinged slightly yellow.

I now prepare the fecal suspension as follows:

A soft portion of the fecal mass is selected and thoroughly triturated in about ten volumes of saline and strained through cheese cloth to remove the coarser particles of fecal debris. The suspension is then centrifuged until the supernatant fluid is clear. If not bile stained and if its reaction is neutral, about one cubic centimeter of the sediment is added to each of a number of test tubes containing about ten cubic centimeters of physiological saline. Without sterilization these are employed for subculturing, one or more being reserved as controls to indicate that flagellates are not present in the feces employed in making the suspension. If the supernatant wash fluid were bile stained it might be possible to secure a satisfactory suspension by one or more washings to remove the excess of bile, but so far I have not had to do this. The suspension thus consists of living fecal bacteria and small particles of undigested or partially digested food. The bacteria prey on the latter and the trichomonads prey on the former. In old cultures there is a very noticeable reduction in both the bacterial and food debris.

Since its cultivation *in vitro* our strain of *Trichomonas* has been carried through subcultures as follows:

No.	Source	Age of Parent Subculture at Time, Days	Greatest Age at which Motile Flagellates Found, Days	Greatest Age at which Subcultures Gave Growth, Days
1	Original	..	59	60
2	1	13	46	47
3	2	18	28	65
4	Failure *	..	..	..
5	3	29	27	..
6	5	24	4	..
6	3	65	9	..
7	6	9	..	..

* Probably due to excess of bile in feces.

From the foregoing it may be seen that these cultures retain their vitality, as measured by the presence of active flagellates, for a period which compares favorably with those of some of the more rugged pathogenic bacteria when growing *in vitro*. Not much growth is evident within the forty-eight hours following a transfer, but it apparently reaches a maximum when the culture is from four to five days old. A loopful of the sediment, placed on a slide and flattened out with a cover glass, may with the low power, reveal nearly a hundred flagellates per field. Following this, their numbers undergo a steady diminution, until not over two or three will be observed in an entire preparation made from a loopful of sediment. I have reason to believe that this diminution in the numbers of flagellate forms is due to encystment.

My observations have convinced me that this organism is *Trichomonas intestinalis,* though as noted before, I have failed to observe an axostyle in the fixed preparations I have made. Nevertheless, an axostyle is distinctly visible in the living organism, projecting posteriorly as a rigid spinous process, as shown in the outline drawings in the plate herewith. The axostyle apparently serves as an organ for temporary attachment. I have observed individuals attached by its assistance to the cover glass or food debris twist and rotate upon their axostyle very much in the manner of a vorticella. When fully active the three anterior flagella move at such a rapid rate they cannot be seen. However, in sluggish individuals they are distinctly visible, and it can be seen that their movement partakes of a rotary character, as indicated in Figure 4. The anterior portion of the undulating membrane is rarely visible during life, but the posterior portion, apparently having a longer membrane, can readily be seen. In very sluggish individuals its movements are suggestive of the movements of pseudopodia, especially where the flagellate is in the pre-encysting stage. From observations of living organisms, it seems probable that the idea these organisms may project pseudopodia, as suggested by Wenyon (1915) may have arisen from observation of the movements

of the posterior portion of the undulating membrane in sluggish individuals. In the pre-encysting state, when the organism, deprived of all appendages, seems to possess a distinct ectoplasmic-endoplasmic differentiation and thus somewhat resembles a small amoeba, the sluggish movements of the undulating membrane are still more suggestive of pseudopodia. However, the ectoplasmic bulgings, representing the altered undulating membrane, first appear toward the anterior extremity and slowly progress posteriorly. These are indicated at the arrows in Figures 7, 8 and 9.

In stained preparations the organism is seen to possess three anterior flagella, and a single posterior flagellum, having an origin at the same place as the others, but projecting posteriorly and forming the free edge of the undulating membrane. It may extend beyond the posterior extremity of the cell body as a free flagellum. A distinct chromatic line at the base of the undulating membrane is visible. All four flagella arise from an anterior blepharoplast or kinetonucleus. The cytostome has not been observed in any of the individuals examined, neither have chromatic blocks been observed about the nucleus. The interior of the cell body invariably contains food vacuoles of varying size, in many of which can be observed bacteria in various stages of digestion. As previously noted, I have not observed the axostyle in fixed specimens, though it is clearly visible in the living animal.

Multiplication appears to be by longitudinal division, an example of which is indicated in Figure 5, in which there can be observed two nuclei, two sets of flagella and two chromatic lines, but cleavage of the cell body has not yet occurred.

In an endeavor to see if the culture strain could re-establish itself in an animal, two white rats were selected, and by fecal examination determined to be free from trichomonads. One rat (A) was given one feeding of milk to which had been added a portion of Culture 2, at that time forty-two days old. Not over one motile flagellate was present per slide preparation, while the culture chiefly contained the spherical bodies later recognized as cysts. The other rat (B) was kept in another cage as a control, receiving similar feed and treatment, but was not fed infected food. Six days later a single motile trichomonad was observed in preparations made from a fecal pellet of Rat A, while similar preparations from Rat B were negative. The entire remaining fecal pellets from both rats were transferred to tubes of saline and separately suspended therein. Three days later these cultures were examined, and from four to five trichomonads were observed per field in the culture from the feces of Rat A, while none were present in the culture from Rat B. Fourteen days after the infected feeding, both

rats were killed. The results of the examination of Rat A (infected) were as follows: No flagellates were observed in the feces, but spherical bodies regarded as cysts were present; the cecum was full of soft pultaceous feces, which was literally swarming with *myriads* of trichomonads; no trichomonads were observed in the small intestine or stomach. Examination of Rat B (control) was entirely negative for trichomonads. The cecal contents of both rats were transferred to saline. When examined three days later, numerous trichomonads were present in the culture from Rat A, but in numbers much less than in the fresh cecal contents. The culture from Rat B was negative. Motile flagellates were still present in the culture from A when examined a month later.

The mucosa lining the cecum of Rat A appeared perfectly normal and throughout the course of the experiment the rat appeared to enjoy unimpaired health.

In studying the growth in cultures I had at first difficulty in recognizing the trichomonas cysts. Various observers have described objects they regarded as trichomonas cysts, a number of which are figured by Lynch, but the chief claim in favor of each appears to be the fact they have been observed in material in which trichomonads were present, and hence were assumed by the observers to be cysts of the observed protozoan. These conflicting views were found confusing. I finally found that by staining the living trichomonads with dilute neutral red solutions and allowing the preparations containing them to undergo gradual desiccation, cyst formation invariably took place and all stages the process could be observed. Figures 7 to 14 represent the process of cyst formation from the flagellate stage in one individual, while Figures 15, 16 and 17 represent typical cysts. The cysts are perfectly spherical and from 5 to 6 micra in diameter. In some a peripherially located nucleus can be discerned. In the drawings the relative thickness of the double contoured wall is somewhat exaggerated. In cultures, without the aid of neutral red solutions to stain the food vacuoles, the cysts can only be made out with difficulty, since their walls are not highly refractile. I am not satisfied I have as yet observed them in fixed films.

It remains to be ascertained whether infection by feeding results from the flagellates or from the cysts. By analogy the latter would seem probable and my single feeding experiment, taking into consideration the paucity of flagellates present, would tend to support this view.

Reference Cited

Wenyon, C. M. 1915.—Observations on the Common Intestinal Protozoa of Man. London Lancet, 2: 1172, Nov. 27.

Explanation of Plate

All figures are free hand drawings

Figs. 1, 2, 3, 4. Unstained living trichomonads. 1, 2, and 3 show various changes of position of a trichomonad which has temporarily attached itself to the coverslip by the tip of its axostyle. Movements consist of a rotation on its long axis as well as lateral flexion. Flagellar movement is too active for visibility. 4, outline of a trichomonad whose movements were sluggish. The three anterior flagella rotate as indicated by the arrows. At (*a*) the movements of the undulating membrane are most distinct. When movement of the membrane temporarily ceases, the change in the organism's shape is indicated by the dotted line.

Figs. 5, 6. From smears fixed in sublimate alcohol and stained with Heidenhain's iron hematoxylin. 5, beginning longitudinal division. Cleavage of the cell body has as yet not taken place. 6, a typical trichomonad.

Figs. 7 to 17. Cyst formation. Living trichomonads from culture stained with dilute neutral red. The food vacuoles stain a dark cherry red and the nucleus appears as a pale pink disk. Figs. 7-14, progressive changes in cyst formation. Changes observed in a single individual. 7. Pre-encysting form. Sluggish, no flagella observable, movements of undulating membrane, indicated by arrow, very sluggish and resemble pseudopodia. 12:20 p. m. 8. Some contraction of the body. Axostyle retracted. Undulating membrane still moving. 12:50 p. m. 9. Distinct differentiation into ecto- and endo-plasm. Undulating membrane moving slightly. 1:15 p. m. 10. Some extension of ectoplasm, approaching original shape. 1:40 p. m. 11. Ectoplasm again contracting. 1:45 p. m. 12. Nucleus no longer visible. Outline nearly spherical. 1:50 p. m. 13. Peripheral ectoplasm becomes denser, outline distinctly double contoured. 1:52 p. m. 14. Perfectly spherical, double contoured wall. Clear zone of ectoplasm has disappeared. Cyst complete. 1:57 p. m.

Figs. 15, 16, 17. Typical cysts. Nucleus in peripheral situation is visible in 15. Food vacuoles visible in all. Diameter from 5 to 6 micra.

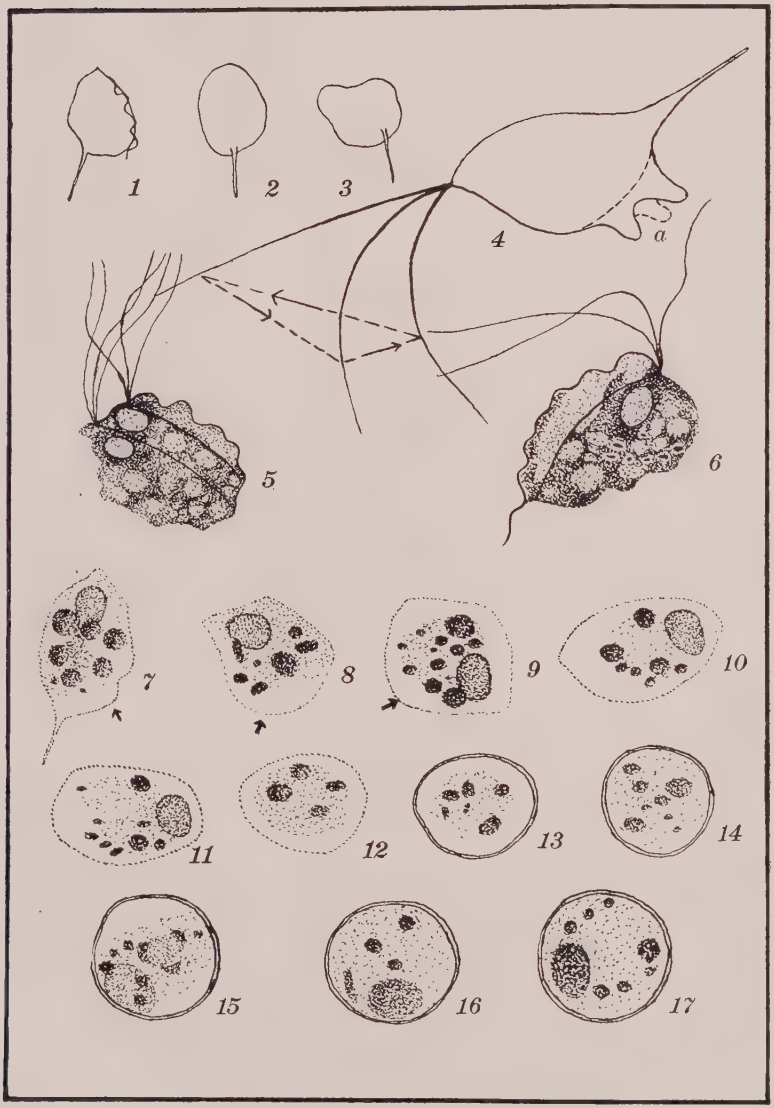


PLATE XI

A CASE OF *BALANTIDIUM COLI* DYSENTERY

C. W. Mason, M.D.
CHIENG RUNG, YUNNAN, CHINA

Human balantidiasis has been considered extremely rare, but I believe is less so than is usually supposed. Strong (1905) was able to find only 125 reported cases. Walker (1913) claims that "In the Philippine Islands, however, parasitizations with this protozoa appears to be relatively prevalent. The first case here (P. I.) was by Strong in 1904. Subsequently a few cases were reported, notably three fatal cases with necropsy by Bowman (1909 and 1911). Willets (1913) found two cases in examination of 400 stools, and I (Walker) found two cases in the examination of 48 stools. Thirteen cases have been observed in the Philippine General Hospital. In the Bilibid Prison, thirty-five cases have been found in the last two and a half years, an average of more than one a month. From March 4 to March 25 of the present year (1913), eight new cases of parasitization with the protozoan were discovered. However, on account of the infrequent appearance of the parasites in the stools of infected persons and the absence of clinical symptoms in many of the cases, it is probable that parasitization with *Balantidium coli* is frequently overlooked in the routine examination of stools."

For a classical and full description of the etiology and pathology I know of nothing equal to this article of Walker. And anyone interested should obtain that article; but I feel that some of his conclusions are important enough to be quoted here.

"*Balantidium coli* was never found entering the tissues through the lesions in 10 parasitized monkeys having a colitis or ulcerations due to bacteria or other causes.

"In those monkeys in which infection took place, the balantidia *entered the tissues through the sound intestinal epithelium.*

"*Balantidium coli* produces bacteriologically sterile abscesses in the submucosa of an infected intestine.

"*Balantidium coli* is the primary etiologic factor in the symptoms and lesions of balantidial dysentery.

"The latency prevalent in balantidiasis of man is due chiefly to the fact that the patient, although parasitized, is not infected with *Balantidium coli,* but in part to the chronicity of the ulcerative process in infected cases.

"*Every person parasitized with Balantidium coli is liable sooner or later to develop balantidial dysentery.*"

In my routine examination of stools I have seen several infections with this parasite, but I was misled by statements in some of the older books that parasitization with this parasite was of no clinical importance, so I made no record of the number of infections noticed. And so far I have only seen this one case in which there was any serious symptoms.

CASE. E. B., male, aged 30, a Danish missionary in Southwestern Yunnan Province, China. He had been living in most unhygienic surroundings, having a Chinese family, including all their pigs, etc., occupying a part of the same house. Flies in enormous numbers swarming on his food at meal time. He had brought his wife to me for confinement. He was a strong, well nourished individual and boasted that he had never taken a dose of medicine. While here he came down with an attack of malaria fever. He was convalescing from this when he began having dysenteric stools. In appearance and odor these seemed to be a typical case, and as amebic dysentery is endemic here I naturally suspected that. He had from twelve to fifteen movements in twenty-four hours. He did not tell me until the third day of his disease, he seemed to be ashamed that he was ill. I then made a microscopical examination of the stools and found them swarming with *Balantidium coli.* I could find no ameba either active or encysted and the specimen seemed almost free of bacteria.

Treatment.—Being at a loss as to a specific against this infection, my mind naturally turned to the weapons used to fight amebic dysentery. At the suggestion of Dr. M. E. Barnes of the International Health Board (Rockefeller Foundation), Chiengmai, I had been trying the oil of chenopedium as an amebacide in amebic dysentery. His suggestion was to give the oil in solution with castor oil by mouth. I had modified it by dissolving it in olive oil and giving 50 to 60 minims in a half ounce of olive oil injected just inside the internal sphincter and I had found that reverse peristalsis soon relieved the desire to go to stool. It seemed logical that as this disease is entirely a disease of the lower bowel the results should be quicker and more efficacious as the drug used would not be subjected to the chemical laboratory actions of the entire alimentary tract and would be in higher concentration. So I gave him an enema of 60 minims oil of chenopodium in half an ounce olive oil. He retained it two hours. The very first movement had fecal matter in it. The second was a formed motion. Next day he was apparently well. Had only two motions both formed and no blood or mucus to the naked eye. The third day I found a few very sluggish Balantidia in his stool and repeated the same enema. Six days later he had a slight relapse but reported it at once and I gave him a third dose. He left me three weeks later with no symptoms and the stools were negative on several examinations. I feel that there is a grave chance of a return of his trouble, but I wish to bring before my colleagues this fact, that he made such a rapid and complete symptomatic recovery and that without any particular restriction of diet except to withhold fruits and coarse rye graham bread for a few days. I fully realize that this was a purely empirical treatment and I pass it on that others may do the scientific experimenting with this drug. The main thing to me or any practitioner was, my patient got well.

PAPERS CITED

Strong, R. P. 1905.—The Clinical and Pathological Significance of Balantidium coli. Bur. Govt. Labs., Manila, Bull. 26.

Walker, E. L. 1913.—Experimental Balantidiasis. Phil. Jour. Sci., 8B: 333-350.

NEW HUMAN PARASITES

THE JOURNAL will endeavor to print very promptly brief reference to all new species of human parasites published since January, 1919. The cooperation of authors and investigators will materially assist in making this of value.

Oxyuris incognita Kofoid and White, 1919. This species of nematode is based on ova found in feces of 1.2 per cent. of about 30,000 soldiers at Camp Travis, Texas, infected individuals coming from 22 states. Adult worms were not discovered. (Jour. Am. Med. Assn., 72: 567, Feb. 22, 1919.)

Leptospira sp. Noguchi, 1919. This protozoan organism, closely resembling that previously found in cases of infectious jaundice, was isolated by Noguchi from guinea pigs inoculated with the blood of yellow fever patients (in 6 out of 27 cases studied at Guayaquil, Ecuador) and apparently the same organism was found in the blood and liver of human patients. It was also found in guinea-pigs inoculated with blood of experimentally infected animals after its passage through a Berkefeld filter. The guinea-pigs showed symptoms and lesions suggestive of those of yellow fever in the human subject. (Jour. Am. Med. Assn., 72: 187, Jan. 18, 1919.)

Dicercomonas soudanensis Chalmers and Pekkola, 1919. This species was discovered in human feces in Khartoum (Sudan) in cases of diarrhea, but is not regarded as pathogenic. The article includes a brief but very important discussion of the classification of the Tetramitidae with the modifications involved by the addition of this new form. (Jour. Trop. Med. Hyg., 22: 29, Feb. 15, 1919.)

Isospora hominis Rivolta, 1878.
Eimeria wenyoni Dobell, 1919.
Eimeria oxyspora Dobell, 1919.
Eimeria (?) sp.

The coccidia parasitic in man are revised by Dobell; he recognizes four species, two of which are new. *I. hominis* appears to be the most common, about 70 definite cases having been recorded. The cases found since 1915 were in men who had been in Egypt, Gallipoli, Salonika or Mesopotamia. *E. wenyoni* has been found four times in persons from the eastern Mediterranean region. *E. oxyspora* has been found only once in a young man who has been in South Africa, Ceylon and India. These three forms are known only from the cysts found in the feces of affected persons, but their habitat is probably the small intestine. The fourth species discovered by Gubler (1858) is the hepatic coccidium of man very imperfectly known, but apparently belonging in the genus *Eimeria*. Since the first case found in Paris four others have been recorded in Prague, Vienna, Giessen and London, none having been reported since 1890. The hepatic parasite is the only one of the four known to be seriously pathogenic. (Parasitol., 11: 147-197; Feb., 1919.)

REVIEW AND NOTES

An extensive and thorough study of the Tsutsugamushi disease has been published in recent numbers of the *Kitasato Archives of Experimental Medicine* by T. Kitashima and M. Miyajima. The concluding section which appeared in December, 1918, includes data on the biological investigations of the virus and a zoological discussion of the mite which acts as intermediate host, and of the field mice which serve as a reservoir of the virus. The authors have reached the positive conclusion that on the basis of the nature of the virus, which is ultra-microscopic, the disease must be classed with the acute infectious diseases among which Spotted Typhus and Rocky Mountain Spotted Fever are to be placed. The number is illustrated by ten splendid plates which include among other things admirable representations of the mite and of the pathology of the disease.

The United States Interdepartmental Social Hygiene Board appropriated from the Scientific Research Fund to Stanford University Medical School, $7,200, and to the University of Michigan, College of Medicine, $6,000 for research work on various problems in venereal diseases. It is in position to make further grants under stated regulations to institutions fitted to carry on such research.

Thirty days after the treaty of peace shall have been signed a congress of Red Cross delegates is to be held in Geneva under the auspices of the International Committee of the Red Cross. The campaign it is proposed to inaugurate at once contemplates a world movement for the prevention of disease as well as for its relief. Parasitology is likely to assume an important place in the discussions of the conference.

The current scientific press records the death, on February 7, of Professor Raphael Blanchard of Paris, the distinguished parasitologist, editor of the *Archives de Parasitologie* and noted for a long series of investigations and publications in this field. The Journal will publish in a later issue an article on Professor Blanchard's life and work.
